PROGRAMA DE DOCTORADO EN FISIOTERAPIA

DETERMINACIÓN MEDIANTE TENSIOMIOGRAFÍA DE LA PRESENCIA DE FATIGA Y LAS DIFERENCIAS ENTRE SEXOS EN LA RIGIDEZ Y VELOCIDAD DE CONTRACCIÓN DE LOS VIENTRES MUSCULARES DEL MUSLO

TESIS DOCTORAL

Presentada por:

Rodrigo Martín De San Agustín

Dirigida por:

Dr. Josep C. Benítez Martínez

Dr. Francesc Medina i Mirapeix

Dr. Jose Casaña Granell

Valencia, Febrero de 2020.

Universitat de València — Facultat de Fisioteràpia

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D. Josep C. Benítez Martínez, Doctor en Fisioterapia por la Universidad Católica San Antonio de Murcia y Contratado Doctor del Departamento de Fisioterapia de Universidad de Valencia.

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CERTIFICAN que el trabajado presentado como Tesis Doctoral por D. Rodrigo Martín San Agustín, titulado **DETERMINACIÓN MEDIANTE TENSIOMIOGRAFÍA DE LA PRESENCIA DE FATIGA Y LAS DIFERENCIAS ENTRE SEXOS EN LA RIGIDEZ Y VELOCIDAD DE CONTRACCIÓN DE LOS VIENTRES MUSCULARES DEL MUSLO,** ha sido realizado bajo nuestra dirección y consideramos que reúne las condiciones apropiadas en cuanto a contenidos y rigor científico para ser presentado a trámite de lectura.

Y para que así conste, expiden y firman la presente certificación en Valencia, a 14 de febrero de 2019.

Fdo.: Josep C. Benítez Martínez
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Fdo.: Josep C. Benítez Martínez
Tutor

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Índice de abreviaturas

TMG: Tensiomiografía

RF: Recto Femoral

VM: Vasto Medial

VL: Vasto Lateral

BF: Bíceps femoral

ST: Semitendinoso

Dm: Máxima deformación radial

Tc: Tiempo de contracción

Td: Tiempo de retardo

VC: Velocidad de contracción al 10-90% de la Dm

V10: Velocidad de contracción al primer 10% de la Dm

Vrn: Velocidad de Respuesta Normalizada

MCIV: Máxima Contracción Isométrica Voluntaria

DE: Desviación Estándar

IC: Intervalo de Confianza

MCD: Mínimo Cambio Detectable

RME: Respuesta Media Estandarizada

ROC: Característica Operativa del Receptor

AUC: Área Debajo de la Curva

Índice

Resumen de la tesis

1. Introducción

El músculo esquelético ha sido extensamente estudiado en las últimas décadas con el fin de conocer tanto su componente anatómico como fisiológico (Sanger & Sanger, 2014). En relación al componente anatómico, su estudio ha abordado por ejemplo la arquitectura muscular tanto en cadáveres (Kumazaki, Ehara, & Sakai, 2012) como en población sana y patológica (Giles, Webster, McClelland, & Cook, 2015; Noorkoiv, Stavnsbo, Aagaard, & Blazevich, 2010). Por otra parte, el examen de la fisiología muscular ha profundizado en las distintas funciones del músculo, entre ellas y como principal, la contracción muscular y los derivados de ésta.

La contracción muscular es la base del rendimiento muscular, entendiéndose éste como la capacidad de un músculo o grupo de músculos para generar fuerzas con el fin de producir, mantener y modificar posturas y movimientos, que son un requisito previo para la actividad funcional (American Physical Therapy Association, 2003). Entre las propiedades del músculo que se relacionan con el rendimiento muscular se encuentran la fuerza, la potencia, la resistencia, el reclutamiento y la longitud (American Physical Therapy Association, 2003). Para evaluar y cuantificar la fuerza, se han propuesto múltiples métodos, clasificados según el tipo de fuerza que evalúan. Para evaluar la fuerza isométrica, existen dinamómetros fijos o de mano que permiten obtener la fuerza en un determinado punto del recorrido articular, y de ese modo, conocer en ese momento angular los kilogramos o newtons generados por la musculatura de la articulación (Bohannon, 1997; Chamorro, Armijo-Olivo, De la Fuente, Fuentes, & Javier Chirosa, 2017). Para movimientos dinámicos, el método generalizado es la cuantificación de la carga desplazada en un movimiento determinado, bien en gestos donde solo intervenga un músculo (Guex, Daucourt, & Borloz, 2015), bien en gestos funcionales donde intervengan varios (Andersen, Vinstrup, Jakobsen, & Sundstrup, 2017). También existen las máquinas isocinéticas, que permiten conocer la fuerza concéntrica o excéntrica que realiza un músculo a una velocidad de movimiento preestablecida (Muff et al., 2016).

Para evaluar la potencia, en función del gesto realizado, se pueden utilizar diferentes herramientas como son los acelerómetros o sensores inerciales, los cuales determinan la potencia conociendo la velocidad de ejecución y la carga movilizada (Orange et al., 2018). En cuanto a la resistencia muscular, los métodos que la evalúan suelen buscar cuantas repeticiones de un determinado gesto es capaz de realizar un músculo en un periodo de tiempo, o bien, cuantas repeticiones dinámicas o cuanto tiempo estático es capaz de aguantar un músculo hasta su fatiga (Friesenbichler et al., 2018; Melchiorri & Rainoldi, 2011). El reclutamiento muscular se refiere a la activación de unidades motoras adicionales para lograr un aumento de la fuerza contráctil en un músculo (Dideriksen, Muceli, Dosen, Laine, & Farina, 2014). La activación muscular evaluada mediante la electromiografía de superficie es el método generalizado que se utiliza para medir el reclutamiento inicial de un músculo al realizar un gesto (Vitry, Martin, Deley, & Papaiordanidou, 2019).

Por último, la longitud muscular hace referencia a la máxima extensibilidad de una unidad músculo-tendón. Esta propiedad ha sido examinada dada la relación entre una óptima longitud muscular con la capacidad de generación de fuerza (Ruas et al., 2019) o con el rango de movimiento (Opplert & Babault, 2019), evaluándose comúnmente por el recorrido articular que se alcanza al llevar a un músculo a su máxima extensibilidad. Además, una propiedad muscular que se podría subclasificar en ésta, es la rigidez muscular. La rigidez muscular se define como el cambio en la resistencia pasiva al alargamiento de un músculo en relación con su cambio en la longitud (Gajdosik, 2001). Mediante el uso de acelerómetros en test dinámicos se puede evaluar globalmente la rigidez muscular de grupos musculares (Kevin P. Granata, Wilson, & Padua, 2002). Para evaluar analíticamente la rigidez de vientres musculares han sido propuestas otras herramientas como el myometer o la tensiomiografía (TMG), que, mediante la deformación radial del músculo, son capaces de medir indirectamente la rigidez muscular (Ditroilo, Hunter, Haslam, & De Vito, 2011; Pisot et al., 2008).

El examen de cada una de estas propiedades con sus respectivos métodos de evaluación ha sido propuesto con diferentes fines y utilidades. En el ámbito del rendimiento deportivo, desde hace décadas las evaluaciones de la fuerza, potencia y resistencia muscular han sido fundamentales a la hora de conocer la condición física del deportista. Para ello, se ha tratado de hacer inferencias y relacionar tales propiedades del músculo con otras que expresan directamente el rendimiento en una tarea, tales como la velocidad en un sprint (Bezodis, Willwacher, & Salo, 2019), la potencia en un salto (McKinlay et al., 2017) o la resistencia en un test de carrera (Ehrström et al., 2018). Del mismo modo, la monitorización de estas propiedades musculares también es utilizada en el deporte para evaluar cambios en éstas y determinar qué tipo de entrenamientos e intervenciones influye en mayor o menor grado en ellas (K. Beattie, Kenny, Lyons, & Carson, 2014; Fieseler et al., 2015; Kildow, Wright, Reh, Jaime, & Doberstein, 2019; Speranza, Gabbett, Greene, Johnston, & Sheppard, 2017). En cuanto al ámbito clínico, la evaluación de las propiedades musculares se han utilizado para conocer la condición del paciente, como por ejemplo, el examen de la fuerza muscular en rehabilitación postquirúrgica (Blackburn & Norcross, 2014) o en patologías articulares (Hilberg, Herbsleb, Puta, Gabriel, & Schramm, 2003; Magalhães, Silva, Sacramento, Martin, & Fukuda, 2013; Topp, Woolley, Hornyak, Khuder, & Kahaleh, 2002). Semejante al rendimiento deportivo, la monitorización de la fuerza o resistencia muscular en el ámbito clínico también ha sido utilizada para observar la progresión de una intervención, en este caso de un tratamiento (Bartholdy et al., 2017; Swank et al., 2011) o fijar un objetivo en un entorno de rehabilitación determinado por valores de referencia (Bohannon, 1997).

Por otra parte, un ámbito que aúna tanto el del rendimiento deportivo como el clínico, es el ámbito de la prevención. La medición de las propiedades del músculo ha adquirido un rol fundamental dentro de los planes de prevención e identificación de factores de riesgo, siendo los cuádriceps e isquiotibiales los dos grupos musculares más analizados (Alentorn-Geli et al., 2015; Fousekis, Tsepis, Poulmedis, Athanasopoulos, & Vagenas, 2011; Hughes & Watkins, 2006; Tam et al., 2017a). Un claro ejemplo de ello es el uso de la medición de la fuerza en diversos deportes como el futbol (Fousekis et al., 2011; Lehance, Binet, Bury, & Croisier, 2009; Nilstad, Andersen, Bahr, Holme, & Steffen, 2014), balonmano (Fieseler et al., 2015; Zebis et al., 2016) o béisbol (Donatelli et al., 2000) para identificar riesgos de lesión. Del mismo modo, la activación muscular durante diferentes test (Baratta et al., 1988; T. E. Hewett, Zazulak, Myer, & Ford, 2005), valores de test de saltos (Heishman et al., 2019) o la evaluación neuromuscular mediante TMG (Alentorn-Geli et al., 2015) han sido también propuestos como métodos evaluativos de factores de riesgo de lesión. Además, dentro de los factores de riesgo que más han sido evaluados en la última década, y de los cuales existe una extensa literatura al respecto, están las diferencias sexuales en distintas funciones musculares y la respuesta de las funciones musculares ante la presencia de fatiga en el deportista.

En relación al sexo, ha sido evidenciado que las diferencias sexuales en la fuerza de cuádriceps o isquiotibiales (Kuenze et al., 2019; Wu, Delahunt, Ditroilo, Lowery, & De Vito, 2016) o en la relación entre estos grupos expresada mediante ratios (El-Ashker, Carson, Ayala, & De Ste Croix, 2017; Timothy E. Hewett, Myer, & Zazulak, 2008) pueden explicar el mayor número de lesiones del miembro inferior en el sexo femenino y un riesgo de lesión mayor. De manera similar, las diferencias sexuales en la activación muscular (Bencke & Zebis, 2011; Ebben et al., 2010; Hannah, Folland, Smith, & Minshull, 2015; Krishnan, Huston, Amendola, & Williams, 2008), rigidez (Blackburn, Riemann, Padua, & Guskiewicz, 2004; Kevin P. Granata et al., 2002; Wang, De Vito, Ditroilo, Fong, & Delahunt, 2015) o velocidad de contracción (D. Rodríguez-Ruiz et al., 2014; David Rodríguez-Ruiz et al., 2012) de los cuadriceps e isquiotibiales también han sido consideradas motivos de los mayores índices lesionales en mujeres. Además, en varias de estas funciones musculares, sus valores han sido normalizados por las características antropométricas de los participantes para compararlos, con el fin de observar si tales características afectan a las diferencias sexuales encontradas en los valores sin normalizar (Blackburn, Bell, Norcross, Hudson, & Kimsey, 2009; Blackburn et al., 2004; Ebben et al., 2010; K. P. Granata, Padua, & Wilson, 2002; Kevin P. Granata et al., 2002; Hannah et al., 2015).

En cuanto a la evaluación de la fatiga y su influencia en las funciones musculares, en primer lugar su presencia ha sido evaluada con diferentes métodos, tanto invasivos como análisis de lactato (Gorostiaga et al., 2012) a otros que requieren de realizar un nuevo esfuerzo en situación de fatiga como test de fuerza (Abbaszadeh-Amirdehi, Khademi-Kalantari, Talebian, Rezasoltani, & Hadian, 2012) o potencia en salto (Raeder et al., 2016). De este modo, la pérdida de fuerza o potencia, cambios en la activación muscular (Garrandes, Colson, Pensini, Seynnes, & Legros, 2007; Thorlund, Michalsik, Madsen, & Aagaard, 2008) o alteraciones biomecánicas (Liederbach, Kremenic, Orishimo, Pappas, & Hagins, 2014; Tam et al., 2017b) son factores que indicarían la presencia de fatiga y un aumento del riesgo de lesión del deportista. Más recientemente, aunque el análisis de su aplicación aun es limitado, se ha evaluado el uso de técnicas que no requieran de un esfuerzo en situación de fatiga (i.e. TMG) para medirla (de Paula Simola et al., 2016; García-Manso et al., 2011).

1.1 Objetivos

Los objetivos generales de esta tesis son los siguientes:

1. Examinar las diferencias entre sexos de la rigidez y velocidad de contracción de 5 vientres del muslo; tres vientres de los cuadriceps [Recto Femoral (RF), Vasto Lateral (VL) y Vasto Medial (VM)] y dos de los isquiotibiales [Bíceps femoral (BF) y Semitendinoso (ST)].
2. Determinar patrones de rigidez y velocidad de contracción y diferencias sexuales entre ellos, estableciendo relaciones entre los 5 vientres y comparando cada patrón entre sexos.
3. Evaluar la sensibilidad al cambio de la TMG a la fatiga local de los cuadriceps.

2. Metodología

2.1. Diseño y sujetos.

Se realizaron dos estudios secuenciales y anidados. Ambos estudios fueron realizados en la Facultad de Fisioterapia de la Universidad de Valencia por estudiantes voluntarios de la misma.

El primer estudio fue un estudio descriptivo transversal en el cual únicamente se evaluaron a los sujetos mediante la TMG en una situación de reposo. Participaron veinte hombres (edad: 21.0 ± 1.97 años, peso: 75.25 ± 10.6 kg y altura: 179 ± 8 cm) y veinte mujeres (edad: 20.5 ± 2.03 años, peso: 54.25 ± 5 kg y altura: 164 ± 6 cm) y el protocolo experimental fue aprobado por el Comité de Ética de la Universidad de Valencia (H1485963491056).

El segundo estudio siguió un diseño pre-post en el cual se realizaron mediciones de TMG y fuerza antes y después de una intervención de fatiga. Treinta y nueve voluntarios (edad: 21.0 ± 1.97 años, peso: 75.25 ± 10.6 kg y altura: 179 ± 0.08 cm) participaron en este estudio. El protocolo experimental fue aprobado por la Comité de Ética de la Universidad de Valencia (H1523633864087).

Existieron criterios comunes de inclusión tales como: (a) edad entre 18 y 30 años, (b) no operados quirúrgicamente de la extremidad inferior, (c) sin dolor del miembro en los 2 meses anteriores a la recolección de datos, (d) realización de ejercicio físico un mínimo de 3 días por semana y (e) capaz de proporcionar consentimiento informado por escrito. Los criterios de exclusión fueron: (a) practicar un deporte específico como amateur o profesional, (b) contraindicación para el uso de electrodos debido a lesión o alergia al adhesivo y (c) no tolerancia a estimulación eléctrica.

2.2. Procedimientos.

2.2.1. Metodología de la TMG

La TMG ha sido desarrollada para valorar la deformación muscular de un vientre tras provocarle una contracción involuntaria, y de ese modo, poder analizar por una parte el componente pasivo al no requerir de voluntariedad del participante para la contracción, y a su vez, examinar el comportamiento contráctil de la misma.

La metodología en la que se basa la TMG consiste en la colocación de un sensor perpendicular al vientre muscular a evaluar con dos electrodos equidistantes a 5 cm del mismo. Así, tras aplicar una estimulación eléctrica, la contracción involuntaria provoca un desplazamiento del sensor. Mediante este sensor conectado a la TMG, se registra la deformidad radial del músculo y distintos parámetros temporales relacionados con la obtención de esa deformidad. En la evaluación tensiomiográfica, el objetivo es conocer la máxima deformación radial del músculo (Dm), la cual ha sido relacionada con la rigidez muscular, al considerarse que ante una mayor deformación ese músculo presenta una rigidez menor (Pisot et al., 2008) y parámetros temporales, los cuales se podrán relacionar con la Dm para conocer aspectos de la velocidad de contracción del músculo. Entre los parámetros temporales de la TMG, el más utilizado es el tiempo de contracción (Tc), el cual es el lapso de tiempo entre el 10% y el 90% de la Dm. Este parámetro ha sido relacionado con el tipo de fibra muscular (rápidas/lentas) (Rusu et al., 2013). Otros parámetros temporales, pero menos utilizados que el Tc son el tiempo de retardo (Td), el cual es el tiempo que tarda la estructura muscular en alcanzar el 10% de la Dm; el tiempo de media-relajación (Tr), el cual es el tiempo en que la respuesta muscular disminuye del 90% al 50% de la Dm; y el tiempo de mantenimiento (Ts), periodo de tiempo que la contracción se mantiene mayor de un 50% de la Dm.

Además, el uso combinado de parámetros espaciales (Dm) con temporales (Tc), permite conocer la velocidad de contracción del músculo producida involuntariamente por la estimulación eléctrica. Así, uno de los parámetros de velocidad de contracción, es la VC, el cual surge de relacionar el Tc con el 80% de la Dm. Otro parámetro es la V10, el cual surge de relacionar el Td con el 10% de la Dm. Por último, y con el fin de poder comparar entre diferentes músculos, existe el parámetro de velocidad de respuesta normalizada (Vrn).

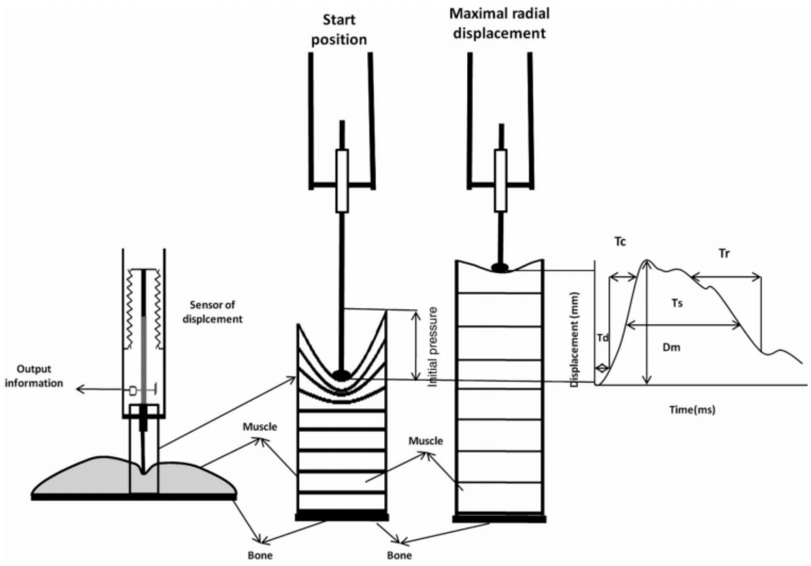

Figura 1. Representación gráfica de la evaluación mediante TMG de un músculo y gráfica obtenida con los distintos parámetros (García-Manso et al., 2011).

2.2.2. Recolección de datos

Los participantes fueron instruidos antes del día de las mediciones de cumplir con las siguientes pautas: (a) no participar en ningún ejercicio vigoroso durante las 48 horas previas a la prueba, (b) no consumir bebidas energéticas o suplementos en las 48 horas anteriores a la prueba, (c) no consumir cafeína o alcohol 3 horas antes de la prueba y (d) no comer alimentos 2 horas antes de la prueba

En primer lugar, se tomaron las características de los participantes mediante una cinta métrica para la estatura y un analizador de composición corporal (Tanita BC 418 MA, Tanita Corp, Tokyo, Japan) para el peso. A continuación, se prepararon las zonas de los vientres musculares de los cuádriceps e isquiotibiales que iban a ser examinados. Para ello, la piel fue rasurada y limpiada con una gasa impregnada en alcohol. Posteriormente, se marcaron mediante un rotulador los puntos donde iba a ser colocado el sensor de la TMG, siguiendo los criterios anatómicos utilizados en previos estudios (J. R. Beattie, 1995; Dahmane, Djordjevic, Simunic, & Valencic, 2005; Rey, Lago-Peñas, & Lago-Ballesteros, 2012; Simunič et al., 2011; Tous-Fajardo et al., 2010). Para la medición de los cuadriceps, los participantes fueron colocados en decúbito supino con una flexión de rodilla de 120º donde 180º es la extensión completa. Para la medición de los isquiotibiales, se puso a los participantes en decúbito prono con la rodilla a 150º de flexión. La flexión en ambas posiciones se estabilizó utilizando un cojín de espuma triangular.

Para realizar la medición por vientre muscular, el sensor fue colocado perpendicularmente al vientre, con dos electrodos puestos a 5 cm del sensor como se muestra en la figura 2 (Alentorn-Geli et al., 2015; Alvarez-Diaz et al., 2016; Loturco et al., 2016; Rey et al., 2012; Seijas et al., 2016). Mediante el electroestimulador propio de la TMG, se aplicaron sucesivas estimulaciones eléctricas partiendo de una intensidad de 50 mA. Fijando 10 segundos de descanso entre estimulaciones para minimizar la fatiga (Krizaj, Simunic, & Zagar, 2008), la medición de cada vientre se finalizó tras conseguir la Dm o alcanzar los 110 mA. Se realizaron dos mediciones por vientre, una para asegurar el buen funcionamiento de la TMG y otra para la adquisición de los datos. Los valores de Dm fueron utilizados como indicador indirecto de la rigidez muscular, donde a mayor Dm, el músculo es considerado menos rígido (Ditroilo et al., 2011; Pisot et al., 2008; D. Rodríguez-Ruiz, Rodríguez-Matoso, Quiroga, Sarmiento, & Silva-Grigoletto, 2011).

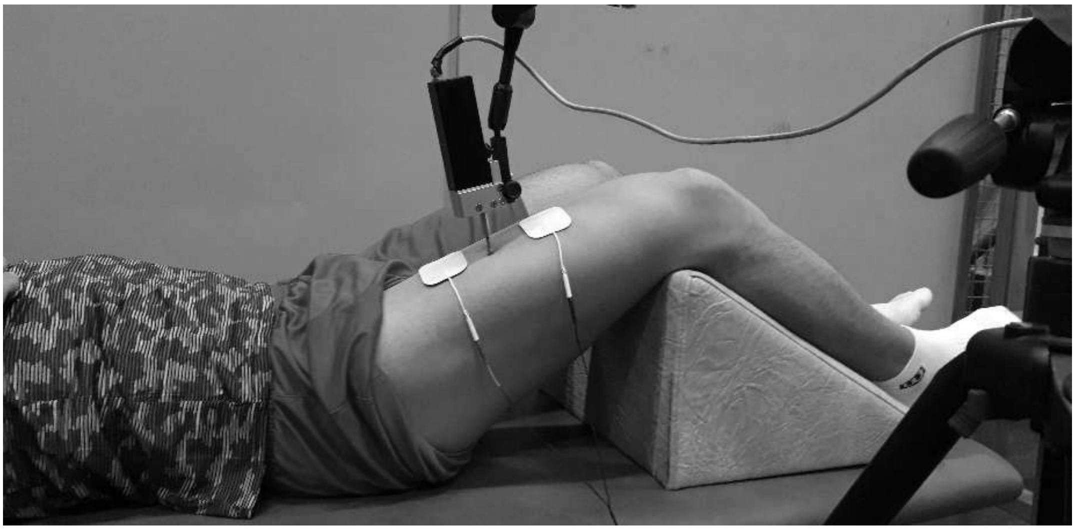

Figura 2. Medición tensiomiográfica del recto femoral.

También se registró el tiempo necesario para obtener la Dm de cada vientre (Simunič et al., 2011; Valencic & Knez, 1997), tanto el Tc como Td, y así, calcular la VC y V10. Además, se calculó la Vrn de cada vientre. Este parámetro relaciona el desplazamiento radial entre el 10 y 90% de la Dm, por el tiempo que se tarda en conseguir (Tc) y es normalizado posteriormente por la Dm, obteniendo un coeficiente de 0.8. Este análisis se explica en las siguientes ecuaciones:

(1) $V_r = \Delta dr/\Delta tc \quad (mm * s^{-1})$

(2) $V_{rn=} V_r/D_m = (\Delta dr/\Delta tc)/D_m \quad (mm * s^{-1}/mm)$

(3) $V_{rn=} 0.8/t_c \quad (mm * s^{-1})$

Este proceso de normalización de la velocidad de contracción ha sido propuesto para poder comparar valores obtenidos entre diferentes músculos (Valencic & Knez, 1997).

A parte de las mediciones con la TMG, en el segundo estudio se realizaron mediciones de la fuerza de los cuadriceps y una intervención para generar fatiga en los mismos.

Para las mediciones de fuerza, se utilizó el dinamómetro de mano MicroFET2 (Hoggan Health Technologies Inc., Salt Lake City, UT). Con los sujetos sentados en un dinamómetro isocinético (Prima Plus, Easytech, Italy) para ser estabilizados, se evaluó la máxima contracción isométrica voluntaria (MCIV) de los cuadriceps a 90º de flexión de rodilla. El dinamómetro manual fue fijado con un cinturón rígido al isocinético y perpendicularmente a la tibia, 2 cm por encima del maléolo y con una almohadilla de foam para minimizar sensación de disconfort sobre la tibia (Hansen, McCartney, Sweeney, Palimenio, & Grindstaff, 2015). Este método ha sido validado previamente, con valores excelentes de fiabilidad (Coeficiente de Correlación Intraclase: 0.93, 95% CI 0.83; 0.97) y un mínimo cambio detectable (MCD) de 14.1 N*m (95% CI, 9.23; 22.01) (Hansen et al., 2015).

Se realizó un calentamiento previo a las mediciones de MCIV que consistió en 5 minutos de bicicleta a ritmo medio y tres contracciones submáximas de extensión de rodilla. Cada participante completó tres MCIV de 5 segundos con 60 segundos de descanso entre repeticiones. Se proporcionó estimulo verbal a los participantes para que realizaran el máximo esfuerzo posible.

Por otra parte, tras realizar las mediciones basales de TMG y MCIV, los participantes realizaron un test de fatiga de los cuadriceps. Este test consistió en mantener durante 60 segundos una contracción mantenida al 70% de la MCIV, utilizando el display del MicroFet2 para dar retroalimentación de la fuerza (Melchiorri & Rainoldi, 2011).

2.3. Análisis estadístico.

Se usó el software SPSS 24 para Windows (SPSS Inc., Chicago, IL, EEUU) para realizar el análisis estadístico. La normalidad de las variables fue comprobada mediante la prueba de Kolmogorov-Smirnov y la homogeneidad de las variaciones con la prueba de Levene. Los valores de Dm y Vrn son presentados mediante sus medias, desviaciones estándar (DE) e intervalos de confianza (IC).

Las diferencias entre sexos de la Dm y Vrn de cada vientre fueron analizadas mediante la prueba T para muestras independientes o Mann-Whitney U (en caso de distribución no normal). También se calculó la *d* de Cohen para evaluar el tamaño del efecto ($d< 0.1$ pequeño, alrededor de 0.3 medio y >0.5 grande). Así mismo, se calculó el porcentaje diferencial entre sexos mediante la ecuación: (Media Dm de mujeres-Media Dm de hombres) / (Media Dm de hombres) ×100. Para la Vrn se calcularon los ratios entre las Vrn de los diferentes vientres en cada sexo. Por último, las diferencias entre sexos de la Dm y Vrn fueron ajustadas por las variables antropométricas (peso y altura) en un modelo de regresión lineal. Se usó el sexo, peso y altura como variables independientes y la Dm o Vrn como variable dependiente. La significación fue establecida en $p<0.05$.

Por otra parte, examinamos relaciones entre la Dm de los diferentes vientres dentro de cada sexo. Mediante la Prueba T para muestras relacionadas, se analizaron diferencias por pares de vientres entre la Dm de todos ellos o entre los ratios calculados para la Vrn. El nivel de aceptación fue elevado a $p<0.01$ al realizarse múltiples comparaciones entre ellos.

En relación al estudio de la sensibilidad al cambio de la TMG a la fatiga, los datos basales se presentaron mediante sus medias y DE para variables continuas, y absolutas y relativas para las categóricas. Se consideró que existió fatiga en los cuadriceps si la reducción en la MCIV fue mayor al límite inferior del MCD de la técnica de medición descrita en previos estudios (22.01 N*m) (Hansen et al., 2015).

Los cambios en los parámetros TMG y MCIV por cada sexo fueron examinados usando una Prueba T para muestras relacionadas. Esos cambios también fueron comparados entre sexos mediante una Prueba T para muestras independientes.

La sensibilidad al cambio interna, que es la habilidad de una medida en cambiar en un periodo de tiempo, fue determinada mediante una Prueba T para muestras relacionadas y el tamaño del efecto (Husted, Cook, Farewell, & Gladman, 2000). La respuesta media estandarizada (RME) fue el estadístico utilizado para analizar el tamaño del efecto, el cual proporciona una estimación de la magnitud de cambio sin estar influenciada por el tamaño de la muestra (Navarro-Pujalte et al., 2018). Los valores de la RME son clasificados en pequeños (0,20-0,50), moderados (0.51-0.80) o grandes (>0.80) (Husted et al., 2000). Además, calculamos el porcentaje de participantes que superaron el MCD.

La sensibilidad al cambio externa, que refleja el cambio de una medida en un periodo de tiempo teniendo como referencia una medida externa, fue examinada mediante el análisis de la correlación entre ambas, modelos de regresión y curvas (ROC) de la característica operativa del receptor (Husted et al., 2000). La referencia de medida externa fue la magnitud de cambio en la MCIV. Las dos primeras pruebas estadísticas fueron utilizadas para relacionar cambios entre los parámetros TMG y cambios en la MCIV, y las curvas ROC fueron utilizadas para discriminar entre participantes fatigados/no fatigados. Un área debajo de la curva (AUC) >0,70 se utilizó como valor para aceptar la habilidad de discriminación de un parámetro TMG para la fatiga (Menaspà, Sassi, & Impellizzeri, 2010).

3. Resultados

Las comparaciones entre sexos de la Dm mostraron en un primer análisis sin ajustar que el BF y RF tuvieron una mayor Dm en mujeres que en hombres, y por ello, una menor rigidez muscular. Sin embargo, en el análisis ajustado por las variables antropométricas, el RF fue el único vientre en mostrar una menor rigidez en las mujeres. Por otra parte, en las comparaciones dentro de cada sexo entre los vientres de los isquiotibiales mostraron en ambos sexos que el ST tenía una Dm mayor que el BF, y por lo cual, una menor rigidez. Dentro de los cuadriceps, el RF mostró ser menos rígido que los vastos en ambos sexos. Por último, los vastos mostraron ser más rígidos que los isquiotibiales en las mujeres y únicamente más rígidos que el ST en los hombres.

Las comparaciones entre sexos de la Vrn sin ajustar mostraron únicamente diferencias entre sexos para el BF, el cual era más lento en las mujeres que en los hombres. En cambio, en el análisis ajustado por peso y altura, el RF fue el único vientre en mostrar diferencias, con valores menores para las mujeres. En relación al análisis de los ratios, el ratio BF/ST presentó diferencias entre sexos, con valores superiores en los hombres frente a las mujeres. En contra, los ratios dentro de los cuadriceps no mostraron diferencias entre sexos. Por otra parte, en los ratios que relacionaron al BF con los tres vientres de los cuadriceps, existieron valores significativos menores en todos ellos en el sexo femenino. Así, en esos tres ratios, las diferencias entre sexos fueron mayores de un 15%.

Por último, el examen de la sensibilidad al cambio de la TMG a la fatiga mostró que, por sexo, la existencia de un patrón de cambio similar en la MCIV y los parámetros TMG, sin diferencias para ninguno de ellos entre sexos. Todos los parámetros TMG, excepto el Tc del RF y VM, tuvieron diferencias significativas entre los valores basales y después del test de fatiga. En relación a la sensibilidad al cambio interna, todos los parámetros TMG, excepto el Tc del RF y VM, mostraron una gran sensibilidad al cambio interna (RME>0,8). La Dm y V10 del RF fueron los parámetros donde más participantes superaron el MCD. Por otra parte, en relación a la sensibilidad al cambio externa, estos dos parámetros, más la Vc fueron los únicos que mostraron relaciones lineales con los cambios de la MCIV. La Dm y V10 del RF también fueron los únicos parámetros que mostraron asociación de sus coeficientes b en modelos ajustados por el género. Por último, varios parámetros TMG mostraron capacidad discriminativa de fatiga (Dm del RF y VL, Tc del VL y V10 del RF y VM).

4. Conclusiones

1. El presente estudio muestra que existen diferencias entre sexos para la rigidez muscular y velocidad de contracción del RF y BF, siendo independientes del peso y la altura las diferencias en el RF.

2. Hombres y mujeres mostraron un patrón similar en la rigidez muscular y velocidad de contracción de los vientres de los cuadriceps e isquiotibiales. Por una parte, los vastos son más rígidos que el RF, y el BF, especialmente en los hombres, que el ST. Por otra parte, la velocidad de contracción de los cuadriceps es mayor que los isquiotibiales, siendo más pronunciada en las mujeres las diferencias entre cuadriceps y BF.

3. Los parámetros Dm y V10 del RF presentan una aceptable sensibilidad al cambio para la fatiga local de los cuadriceps.

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Anexos

Categorización de los artículos presentados en la tesis doctoral

Artículo	Revista	Factor de impacto (según JCR)	Área temática	Ranking	Cuartil
I	Journal of Strength and Conditioning Research	3.017	Sport sciences	18/83	Q1
II	Journal of Strength and Conditioning Research	3.017	Sport sciences	18/83	Q1
III	PeerJ	2.353	Multidisciplinary sciences	27/69	Q2

JCR: Journal Citation Reports (2018).

ARTÍCULO I

Martín-San Agustín, R., Benítez-Martínez, J. C., Medina-Mirapeix, F., & Casaña-Granell, J. (2018). Sex Differences and Patterns of Muscle Stiffness in the Knee Flexor and Extensor Musculature Through Analysis of Isolated Bellies. *Journal of Strength and Conditioning Research*. doi: 10.1519/JSC.0000000000002883

Sex Differences and Patterns of Muscle Stiffness in the Knee Flexor and Extensor Musculature Through Analysis of Isolated Bellies

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Abstract

Martín-San Agustín, R, Benítez-Martínez, JC, Medina-Mirapeix, F, and Casaña-Granell, J. Sex differences and patterns of muscle stiffness in the knee flexor and extensor musculature through analysis of isolated bellies. *J Strength Cond Res* XX (X): 000–000, 2018—Muscle stiffness (MS) is one of the key factors in joint control. The purpose of this study was to determine sex differences in the MS of 5 isolated muscle bellies (biceps femoris [BF], semitendinosus [ST], rectus femoris [RF], vastus medialis [VM], and vastus lateralis [VL]) and in the pattern of differences among their respective MS. Twenty female and 20 male recreational athletes participated. Muscle stiffness was measured by tensiomyography using maximum radial deformation (Dm) as an indirect indicator of MS. Sex differences were observed only in the Dm of RF (mean difference = 2.07 mm, $p < 0.05$) when values were adjusted by body mass and stature. Males and females showed a similar pattern in the Dm between the muscle bellies: within the hamstrings, ST had a significantly higher Dm than BF in females (3.02 mm) and males (4.28 mm); within the quadriceps, RF also had a significantly higher value than VL and VM in females (6.50 and 7.38 mm, respectively) and males (4.87 and 4.82 mm, respectively). Sex differences in patterns were found between BF and the vastus muscles: the BF of females had a significantly higher Dm than VL (3.78 mm) and VM (4.51 mm), but this was not observed in males. Differences may imply different involvement of the bellies in countering the movements of the lower extremities. Our results can help to direct exercises to improve the MS in certain muscular bellies.

Key Words muscular stiffness, knee joint, tensiomyography

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Introduction

Muscle stiffness (MS) is defined as the change in passive resistance to the lengthening of a muscle in relation to its change in length (13). A number of recent reports suggest there are sex differences in the MS of human skeletal muscle (7). In general, females are less rigid than males with regard to the quadriceps femoral (QF), hamstring (HM) (14,26), and the lateral gastrocnemius (21). Considerable evidence exists in relation to factors that explain sex differences in MS (4). These factors are mostly related to morphological aspects, such as muscular mass (32), the presence of stable cross-bridges within the muscle (23), the amount of connective tissue (24) and titin (13,31), and also the geometry of the muscle and its cross-sectional area (12). These sex differences are thought to be a potential factor contributing to the higher risk of knee injury in females when compared with males (14,31). In this sense, it has been suggested that the lower MS of females may lead to alterations in knee joint stability because of decreased potential of the HM as a protective mechanism of the anterior cruciate ligament when countering the anterior displacement of the tibia (3).

Research on sex differences in MS have been focused on the QF and HM because of their importance in control of the knee. Usually, MS measurements of these 2 muscle groups have been taken globally on all of their muscle bellies as a whole by means of an accelerometer during a flexion and extension test (14). However, this methodological approach does not provide analytical information about the MS of muscle bellies. Although there are several instruments that allow for isolated MS measurements (e.g., myometer, tensiomyography [TMG]) (10,22,26), there is few literature that analyzes differences between sexes in isolated muscle bellies (26,31). These instruments measure radial deformation as an indirect indicator of MS (8,18,26). Thus,

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for example, TMG uses a displacement sensor to measure the maximum radial deformation (Dm) of the muscle belly, with greater values representing a decrease in MS (22).

To the best of our knowledge, only 2 studies have compared the MS of isolated muscle bellies of the thigh between sexes (26,31). Nevertheless, these studies analyzed either the biceps femoris (BF) (26) or vastus lateralis (VL) (31) and therefore, we do not know whether there are sex differences in the MS of other isolated bellies or in the pattern of differences between the MS of those bellies.

To investigate these issues, we examined the MS of 5 isolated muscle bellies from HM (BF and semitendinosus [ST]) and QF (rectus femoris [RF], vastus medialis [VM], and VL) using TMG in females and males of similar age and physical activity level. The purpose of this study was to determine sex differences in the MS of 5 isolated muscles bellies (BF, ST, RF, VM, and VL) and in the pattern of differences among their respective MS. With this information, strength and conditioning programs could be more precise to manage MS differences and thus the purpose of injury prevention strategies. We hypothesized that there are sex differences in the MS of isolated bellies, but that they decrease when adjusted for body mass and stature. In this line, we also hypothesized that females and males have a similar pattern of differences between the MS of bellies from HM and QF.

Methods

Experimental Approach to the Problem

We used TMG to measure Dm of the muscle belly and used this parameter as an indirect indicator of MS, where a higher mean Dm represents less MS (22). Participants were instructed before taking measurements to comply with the following guidelines (25): (a) not to participate in any strenuous exercise in the 48 hours before the test, (b) not to consume any energy drink or supplement 48 hours before the test, (c) not to consume caffeine or alcohol 3 hours before test, and (d) not to consume food 2 hours before the test. Before the measurements, the participants were asked if they had fulfilled all the guidelines.

Subjects

Twenty female (mean ± *SD*: age: 20.5 ± 2.03 years, body mass: 54.25 ± 5.0 kg, and stature: 164 ± 0.06 cm) and 20 male (age: 21.0 ± 1.97 years, body mass: 75.25 ± 10.6 kg, and stature: 179 ± 0.08 cm) recreational athletes were evaluated, all of them volunteers from the University of Valencia. All participants performed exercise ~3 times per week and practiced activities such as running, swimming, cycling, or general strength training. The specific inclusion criteria were: (a) aged between 18 and 30 years, (b) not surgically operated on the lower limb, (c) without pain in the lower limb in the 2 months before data collection, (d) performing physical exercise a minimum of 2 days per week, and (e) able to provide written informed consent. The exclusion criteria were: (a) practicing a specific sport as an amateur or professional, (b) contraindication to the use of electrodes due to injury or allergy to the adhesive, and (c) nontolerance to electrical stimulation.

Participants were physiotherapy students recruited by mail using the internal network of the University of Valencia. Before being included, they were informed about possible risks and benefits of the project and signed their informed consent. The experimental protocol was approved by the Ethics Committee of the University of Valencia (Spain) (H1485963491056). Data collection was cross-sectional and performed between February and March of 2017 in the clinical research laboratory of the Department of Physiotherapy (University of Valencia).

Procedures

First, with the participants at rest on a stretcher and the skin of the area to be tested shaved and cleaned with alcohol-soaked gauze, the position of the TMG sensor and of the respective electrodes were marked. The positioning of the sensor was determined following the anatomical criteria used in previous studies (8,9,25,28,29), and the position of the 2 self-adhesive electrodes was symmetrically equidistant at a distance of 5 cm from the sensor, the anode and, distally, the cathode.

The measurement protocol measured muscular radial deformation according to the procedure that has been used previously by several authors (1,2,20,25,27). The measurements were performed with the muscles at rest, in supine position with the knee held at 120° of flexion (with 180° being complete extension) for the muscle bellies of the QF (Figure 1), and in prone position with 150° of flexion for the HM, maintaining a fixed angle with a triangular foam cushion. Muscular radial deformation was measured perpendicular to the muscle belly with a Trans-Tek Dc-Dc digital

Figure 1. Measurement by tensiomyography of the rectus femoris (belly of the quadriceps femoris).

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TABLE 1. Maximal radial deformation of each of the 5 muscles by sex and differences between groups.*†

	Females		Males		Sex differences (females − males)	
					Mean (95% CI); effect size‡	
Muscle	Mean ± *SD*	95% CI	Mean ± *SD*	95% CI	Unadjusted	Adjusted§
BF	8.98 ± 2.97	7.55/10.42	6.66 ± 3.75	4.91/8.42	2.3 (0.14/4.43); 0.68‖	0.52 (−2.71/3.74)
ST	11.96 ± 3.23	10.40/13.52	10.95 ± 2.63	9.72/12.18	1.03 (−0.8/2.87); 0.34	−1.72 (−4.67/1.22)
RF	11.67 ± 1.74	11.07/12.62	9.82 ± 2.01	8.88/10.76	1.86 (0.65/3.06); 0.98¶	2.07 (0.08/4.06)‖
VL	5.18 ± 1.63	4.39/5.97	4.95 ± 1.66	4.17/5.73	0.23 (−0.81/1.26); 0.14	0.13 (−1.53/1.79)
VM	4.47 ± 1.00	3.99/4.95	5.00 ± 1.34	4.37/5.62	−0.52 (−1.29/0.24); −0.44	0.60 (−0.59/1.79)

*CI = confidence interval; BF = biceps femoris; ST = semitendinosus; RF = rectus femoris; VL = vastus lateralis; VM = vastus medialis.
†All values in mm.
‡Effect size was estimated with Cohen's *d* only for the unadjusted model.
§Adjusted by body mass and stature.
‖Significant differences at $p < 0.05$.
¶Significant differences at $p < 0.01$.

transducer (GK 40; Panoptik d.o.o., Ljubljana, Slovenia) at the positions marked previously.

Electrical electrostimulation, applied by a TMG-100 System (TMG-BMC doo, Ljubljana, Slovenia), was performed with a pulse of 1 ms and with an initial amplitude of 50 mA, increasing progressively by 10 mA until there was no increase in the Dm or until reaching 110 mA as previously described (1). This maximum response was typically achieved between 40 and 70 mA (17). Rest periods of 10 seconds were guided between consecutive measurements to minimize fatigue or potentiation effects (18). Two measurements were performed per muscle, one to verify the operation of the TMG and the second as data acquisition for the study.

Statistical Analyses

Participant characteristics are presented as mean values and *SD*s. Muscle deformity measurements are presented as mean, *SD*, and 95% confidence intervals (CIs). Variables were checked for normality with the Kolmogorov-Smirnov test and for homogeneity of variances with Levene's test.

To study sex differences in each muscle belly, unpaired *t*-tests were used to compare deformity mean differences between females and males. Cohen's *d* was also calculated to evaluate the effect size ($d < 0.1$ small, around 0.3 medium, and >0.5 large). In addition, for each muscle belly with significant differences, we calculated the percentage increase or decrease between sex groups using the following equation: $\frac{\text{Mean Dm from women} - \text{Mean Dm from men}}{\text{Mean Dm from men}} \times 100$. Finally, we also compared mean differences adjusted by anthropometric variables (body mass and stature) by linear regression models using sex, body mass, and stature as independent variables and muscle deformity as a dependent variable. Significance tests were 2-tailed, and α was set at 0.05.

In a final step, we explored the relationships between muscles bellies within each sex group. Within each sex, paired *t*-tests were used to assess deformity differences between multiple pairs of muscle bellies and 99% CIs were calculated. To reduce the probability of getting false positives, we increased the acceptance level from 0.05 to 0.01 for these tests because multiple comparisons were made on the same data set.

Results

Sex Differences in Individual Muscle Bellies

Deformity measurements for each muscle belly are listed in Table 1. The unadjusted analyses showed that 2 of the 5 muscle bellies (BF and RF) had a mean MS that was significantly different in the 2 sex groups. Females had a higher Dm than males in these 2 bellies. Thus, BF and RF from females had 34.53 and 18.94% greater mean deformity, respectively. However, when comparisons were made with the mean values adjusted for body mass and stature, only RF had significant sex differences.

Patterns of Relationship Between Muscle Bellies

Table 2 outlines multiple deformity mean ± *SD* differences from paired muscle bellies within and between HM and QF muscle groups. Comparisons within HM bellies show that the mean muscle deformity of ST was significantly higher than BF in both females (3.02 mm) and males (4.28 mm). Within QF, RF deformity was also significantly higher than VL and VM in females (6.50 and 7.38 mm, respectively) and males (4.87 and 4.82 mm, respectively), but there were no differences between vastus muscles.

Comparisons between muscle bellies of HM and QF show similar patterns of deformity relationships between sex

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TABLE 2. Maximal radial deformation differences between paired muscle bellies for each sex group.*†

	Females			Males		
	Mean ± *SD*	99% CI	Effect size	Mean ± *SD*	99% CI	Effect size
Within hamstrings						
ST–BF	3.02 ± 3.32‡	0.89/5.14	0.96	4.28 ± 3.85‡	1.82/6.75	1.32
Within quadriceps						
RF–VL	6.50 ± 2.08‡	5.17/7.83	3.85	4.87 ± 2.42‡	3.32/6.42	2.64
RF–VM	7.38 ± 1.93‡	6.10/8.65	5.07	4.82 ± 2.20‡	3.41/6.23	2.82
VL–VM	0.71 ± 1.58	−0.35/1.76	0.52	−0.04 ± 1.96	−1.30/−1.20	−0.03
Between hamstring and quadriceps bellies						
ST–RF	0.29 ± 3.24	−1.78/2.36	0.11	1.12 ± 3.32	−1.01/1.20	0.48
ST–VL	6.79 ± 3.00‡	4.87/8.72	2.65	5.99 ± 3.25‡	3.92/8.07	2.73
ST–VM	7.49 ± 2.99‡	5.51/9.47	3.13	5.95 ± 3.21‡	3.89/8.00	2.85
BF–RF	−2.72 ± 3.34‡	−4.86/−0.59	−1.10	−3.16 ± 3.83‡	−5.61/−0.71	−1.05
BF–VL	3.78 ± 3.45‡	1.57/5.98	1.58	1.71 ± 3.94	−0.81/4.22	0.59
BF–VM	4.51 ± 2.75‡	2.69/6.33	2.03	1.66 ± 4.42	−1.16/4.49	0.59

*CI = confidence interval; ST = semitendinosus; BF = biceps femoris; RF = rectus femoris; VL = vastus lateralis; VM = vastus medialis.
†All values in mm.
‡Significant differences between paired muscle bellies at $p < 0.01$.

groups, except between BF and vastus muscles. Thus, while the female BF had a significantly higher deformity than VL (3.78 mm) and VM (4.51 mm), the male BF did not. Common patterns were mean muscle deformity of ST and RF was significantly higher than vastus and BF, respectively.

DISCUSSION

Our study examined sex differences in the MS of 5 muscle bellies of the thigh, 2 for the HM and 3 for the QF. We found that females are less rigid in RF, but no statistically significant differences were found for ST, VL, VM, or BF, especially when data were adjusted by anthropometric variables (body mass and stature). In addition, our study evidenced a similar pattern between muscle bellies in males and females, having a greater MS in the vastus compared with the RF, ST, and BF. However, females showed a more pronounced difference in MS between the BF and the vastus than males.

To the best of our knowledge, our study is the first to adjust MS comparisons between HM and QF bellies for body mass and stature. Most previous studies using either global or analytical measurements of HM and QF (14,31) did not adjust their results based on anthropometric data. There are few studies using global measurements that have considered anthropometric variables in MS differences between sexes. Those which did consider them evaluated MS dynamically, either with the applied moment in HM (product of system mass and shank segment length) (4,5) or considering body mass in functional tasks in the leg as a single element (15).

Comparisons of the MS of BF between sex groups show different results between adjusted and unadjusted analyses, with there being no differences when taking into account the anthropometric data. In addition, BF and ST show lower MS differences between sexes in the adjusted analyses than in the nonadjusted ones. Those findings are expected, given previous studies (4,5,14) that showed that differences in MS of HM between sexes disappear when considering body mass. The finding of sex differences for the RF after adjusting for body mass and stature suggests that its MS is less dependent on anthropometric characteristics. Given the high influence that RF has on the joint, this lower MS in females could be a potential risk factor for knee injury.

The unadjusted differences between sex groups for BF and RF are consistent with previous studies because females have lower stiffness than males (14,26,31). Even so, it was an unexpected finding that only BF and RF show differences, and not the rest of the bellies. Previous authors (30) obtained differences in the MS of VL between sexes, using VL as a representative muscle of the QF and Myoton-3 as evaluation tool. Thus, although both techniques (Myoton-3 and TMG) have proved valid for studying changes in MS (10), the different methodology used may explain the differences. Therefore, because Myoton-3 is applied to a muscle at rest and TMG causes a voluntary contraction to measure MS, we believe it is appropriate to normalize their values by a morphological property of the muscle (e.g., cross-sectional area) to compare results between the 2 techniques.

Patterns of differences among the bellies within HM and within QF were similar between females and males.

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Thus, BF and the vastus (VL and VM) have higher MS than ST and RF, respectively. Because each muscular belly has its own structure and function, the characteristic morphology of each could explain these patterns. On the one hand, within the HM, the shorter length of the muscle fibers of the long head of the BF, which has a hemipennate architecture, compared with the ST (with a fusiform architecture), could explain the presence of these differences between them (19). On the other hand, within the QF, the vastus muscles have a higher amount of connective tissue because they have aponeurotic connections with other muscles (16), and this could lead to an increase in their MS.

Our pattern for males among all 5 bellies was similar to that obtained by other authors (12). They also found that the vastus muscles were more rigid than other bellies (BF and RF). However, in females, the authors did not obtain differences between bellies. This difference with respect to our study could be attributed to the influence of physical exercise and strength on stiffness (15) because their sample was composed of elite volleyball players, with the specific work of the thigh that this entails.

To the best of our knowledge, no previous studies have analyzed MS sex differences in both isolated muscle bellies and the patterns between them. Furthermore, the use of a standardized measuring technique and objective measurements that allow for comparisons with other studies could be considered strengths of the study.

Our study had several limitations. We used a technique that measures deformity and used this as an indirect indicator of stiffness, although this technique has been previously used for this, being accepted as a tool to measure stiffness (10). Although anthropometric data were regarded in MS comparisons between sexes, we believe that normalization by the characteristics of each individual muscle would be more appropriate, given their varied morphology. Thus, future research should normalize MS values using mass, volume, or cross-sectional area of each muscle or, alternatively, regarding length of the segments (e.g., femur). In addition, we did not control the menstrual cycle in females and, although the effects of menstruation on MS are controversial (6), the amount of estrogen does seem to affect it (11).

Practical Applications

This study on MS of the thigh in recreational athletes revealed that males and females showed a similar pattern between muscle bellies, having a greater MS in the vastus compared with RF and in the BF compared with ST. Thus, our study suggests that BF will provide greater resistance to elongation than ST, particularly in males, being able to contribute to the countering of tibial anterior displacement to a greater degree. Therefore, this finding may help to guide MS-building exercises and direct them to specific muscle bellies.

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ARTÍCULO II

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Sex Differences in the Velocity of Muscle Contraction of the Hamstring and Quadriceps Among Recreationally Active Young Adults

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Abstract

Martín-San Agustín, R, Medina-Mirapeix, F, Alakhdar, Y, and Benítez-Martínez, JC. Sex differences in the velocity of muscle contraction of the hamstring and quadriceps among recreationally active young adults. *J Strength Cond Res* XX(X): 000–000, 2019—This study determines sex differences in the velocity of contraction (VC) of 5 isolated muscles (biceps femoris, semitendinosus, rectus femoris, vastus medialis, and vastus lateralis) and in the relationships between them. Thirty-six female and 34 male recreationally active young adults participated in the study. The VC was measured by tensiomyography using normalized response velocity (Vrn) to perform comparisons. Sex comparisons were adjusted by height and mass. The study of relationships was carried out by comparing and calculating means and ratios. Sex differences were observed in the VC of rectus femoris (mean difference = 6.20 mm·s^{-1}; $p < 0.001$). Conversely, the biceps femoris only showed sex differences in the unadjusted analysis (mean difference = 6.66 mm·s^{-1}; $p = 0.002$; $d = 0.73$. Both sexes showed lower VC values of the hamstring with respect to the quadriceps. Female participants showed differences greater than 15% relative to male participants between biceps femoris and quadriceps ratios and in ratios in the hamstring. Thus, our findings in the VC ratios indicate different mechanical contractile properties between sexes in the relations between the hamstring and quadriceps. Our analysis of the VC at these muscles supposes a new possibility to establish the relationships between knee agonists and antagonists, which allow monitoring the changes in the balance of the VC among the muscle groups.

Key Words: velocity of contraction, tensiomyography, knee ratio, muscular properties

Introduction

Epidemiologic studies have consistently demonstrated that female individuals have an increased risk of lower limb musculoskeletal sports-related injuries compared with male individuals (26) and also that the incidence of anterior cruciate ligament (ACL) injuries in female individuals is 2.1 greater compared with male individuals, given the same time of exposure and adjusted to the type of sport (6,14,19). Because hamstring forces act as synergists of the ACL against the anterior displacement of the tibia (10,27), sex differences in the balance of muscular activation (17), strength (16,18), stiffness (7), and the velocity of contraction (VC) (30) between the hamstring and quadriceps have been suggested as the reasons for differential rates of incidence of ACL injury (21).

Studies analyzing sex differences in the relationship between the hamstring and quadriceps regarding strength (16) and muscle activation (11) have evidenced that the hamstring has substantially lower strength and muscle activation than the quadriceps and that this difference is more pronounced in female individuals. Thus, the hamstring/quadriceps ratio in female individuals is lower than that in male individuals when this is described using isokinetic strength (51.0 vs. 81.4%) at high knee flexion/extension angular velocities (approaching those that occur during sports activities) and EMG (38 vs. 55%) characteristics. Studies analyzing sex differences in isolated muscles from the hamstring and quadriceps have also been conducted in regard to muscle activation or the VC. These studies have shown that the values from the hamstring are lower in both sexes in muscle activation (ranging between 57 and 85%) and VC (ranging between 8 and 42%) than those from the quadriceps (1,11,30) and that female individuals also have more pronounced differences than male individuals (11,30), especially between biceps femoris and quadriceps.

Most studies on the differences between the VC have been done previously (29,30) using tensiomyography (TMG). Tensiomyography serves as a noninvasive method that can assess the VC of superficial muscles when activated by an electrical stimulus of controlled intensity (29). The analysis of the radial VC produced by an isolated stimulation allows to compare the mechanical contractile properties of different muscles and to observe whether a certain functional demand alters their relationships (29). These studies analyzed and compared the patterns of the VC between biceps femoris and quadriceps (rectus femoris, vastus lateralis, and vastus medialis) in both male and female elite volleyball players (30) and identified sex differences in the VC of some of these muscles (29). However, to date, it remains unknown whether their relevant results are equivalent in nonspecific sport populations (i.e., recreationally active young adults). Moreover, because the study of VC differences between the quadriceps and hamstring has been explored only with the biceps femoris, it still remains uncompleted and not enough known. Similarly, the comparison of the hamstring/quadriceps ratio between male and female individuals remains largely untested.

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To investigate these issues, we examined the VC of 5 isolated muscles using TMG from the hamstring (biceps femoris and semitendinosus) and the quadriceps (rectus femoris, vastus medialis, and vastus lateralis) in female and male recreationally active young adults. This study determines sex differences in the VC of 5 isolated muscles and in the relationships of them. We hypothesized that female individuals have a lower ratio between the hamstring and quadriceps than male individuals.

Methods

Experimental Approach to the Problem

A single day of testing was used to measure the VC of the hamstring and quadriceps in both female and male individuals, to compare the VC between the pairs of agonist and/or antagonist muscles, and to examine sex differences in both isolated muscles and their relationships.

Subjects

Thirty-six female individuals (mean age: 21.1 ± 2.3 years; body mass: 55.9 ± 5.9 kg; stature: 164.8 ± 5.9 cm; weekly physical activity: 224.3 ± 128.8 minutes) and 34 male individuals (mean age: 21.6 ± 2.4 years; body mass: 75.8 ± 10.4 kg; stature: 179.5 ± 7.4 cm; weekly physical activity: 289.2 ± 166.6 minutes) who were recreationally active (engaging in 1–5 hours of moderate physical activity 3–4 days per week) (4) were evaluated; all of them were volunteers from the University of Valencia. Subject characteristics were measured standard deviation. All participants practiced recreational sports such as running, swimming, cycling, or general strength training. The specific inclusion criteria were the following: (a) aged between 18 and 30 years, (b) not surgically operated on the lower limb, (c) without pain in the lower limb in the 2 months before data collection, (d) performing physical exercise for a minimum of 3 days per week, and (e) able to provide written informed consent. The exclusion criteria were the following: (a) practicing a specific sport as an amateur or a professional, (b) contraindication to the use of electrodes due to injury or allergy to the adhesive, and (c) nontolerance to electrical stimulation.

Participants were physiotherapy students recruited by email using the University of Valencia Intranet. Before being included, they were informed about the possible risks and benefits of the project and signed their informed consent. The experimental protocol was approved by the Ethics Committee of the University of Valencia (Spain) (H1485963491056). Before the measurements began, the protocol followed was sent for approval to the Institutional Research Ethics Committee for testing human participants. Data collection was carried out in the clinical research laboratory of the Department of Physiotherapy (University of Valencia).

Procedures

Participants were instructed before taking measurements to comply with the following guidelines (28): (a) not to participate in any strenuous exercise during the 48 hours before the test; (b) not to consume energy drinks or supplements in the 48 hours before the test; (c) not to consume caffeine or alcohol 3 hours before the test, and (d) not to eat food 2 hours before the test.

First, with the participants at rest on a stretcher, the area for the tests was shaved and cleaned with alcohol-soaked gauze; the positions of the TMG sensor and the respective electrodes were marked. The positioning of the sensor was determined following the anatomic criteria used in previous studies (5,9,28,32,33), and the position of the 2 self-adhesive electrodes was symmetrically equidistant at a distance of 5 cm from the sensor, the anode proximally and the cathode distally.

The measurement protocol measured the muscular radial deformation according to the procedure previously used by several authors (2,3,24,25,28,31). The measurements were performed after 5–8 minutes of the participant resting on the stretcher and the usual joint angles to evaluate that musculature (12), in supine position with the knee held at 120° of flexion (with 180° being complete extension) for the quadriceps, and in prone position with 150° of flexion for the hamstring, maintaining a fixed angle with a triangular foam cushion (Figure 1). The muscular radial deformation was measured perpendicular to the muscle with a Trans-Tek Dc-Dc digital transducer (GK 40; Panoptik d.o.o., Ljubljana, Slovenia) at the positions previously marked.

Electrical stimulation on each muscle was performed with a pulse of 1 ms and with an initial amplitude of 50 mA by the application of a TMG-100 System (TMG-BMC doo, Ljubljana, Slovenia), increasing progressively by 10 mA until there was no increase in the maximal radial deformation (Dm) or until reaching 110 mA. This maximum response typically was achieved between 40 and 70 mA (20). Rest periods of 10 seconds were given between consecutive measurements to minimize fatigue or potentiation effects (23). Two measurements were performed per muscle in the same session: the first one to verify the operation of the TMG and the second to acquire data for the study.

As recommended, our protocol recorded the time in which each muscular radial displacement occurred to calculate different parameters of the VC (32,34). In all of them, our study used the normalized response velocity (Vrn). This parameter relates the radial displacement between 10 and 90% of the Dm of the muscle (Δd_r) and the increase in the muscular contraction time (Δt_c) in this displacement (equation 1). This is done by dividing equation 1 by the Dm for each muscle (equation 2). The authors report that (Δd_r) is equal to 0.8 per Dm. Thus, the Vrn would be equal to 0.8 divided by the muscle contraction time (tc) between 10 and 90% (equation 3).

$$V_r = \Delta dr/\Delta tc\left(mm \times s^{-1}\right). \quad (1)$$

$$V_{rn} = V_r/D_m = (\Delta dr/\Delta tc)/D_m\left(mm \times s^{-1}/mm\right). \quad (2)$$

$$V_{rn} = 0.8/t_c\left(mm \times s^{-1}\right). \quad (3)$$

The authors propose the normalization of this increase over time to compare the values obtained in different muscles (34). Other studies also suggested that the calculation of the Vrn makes it possible to isolate the interference that can occur due to the individual characteristics of each participant studied and also the anatomic and functional differences in different muscles (29).

Statistical Analyses

Data were summarized as means (*SD*), and 95% confidence intervals (CIs) were used for Vrn measurements. Variables were checked for normality with the Shapiro-Wilk test and for homogeneity of variances with the Levene test.

To study sex differences in each muscle, unpaired *t*-tests or Mann-Whitney *U* tests (if non-normally distributed data) were used to compare Vrn mean differences between female and male individuals. Cohen's d was also calculated to evaluate the effect size ($d < 0.2$: trivial, 0.2–0.5: small, 0.5–0.8: medium, and >0.8: large) (8). Cohen's $d \geq 0.5$ was considered to be a practically

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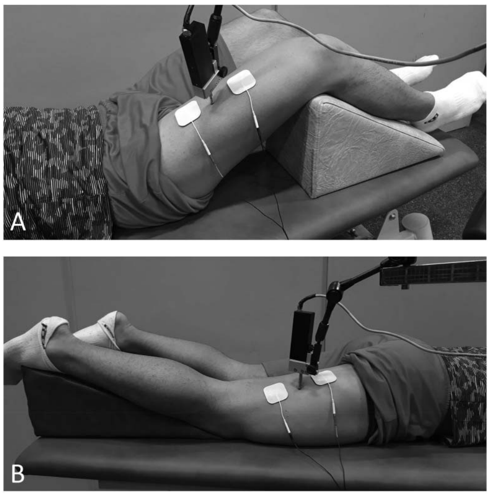

Figure 1. Measurement by tensiomyography of the rectus femoris (A) and the biceps femoris (B).

important difference. Finally, we also compared the mean differences adjusted by anthropometric variables (body mass and height) by linear regression models using sex and anthropometric measures as independent variables and muscle Vrn as a dependent variable. Significance tests were two-tailed, and α was set at 0.05.

In the next step, we explored Vrn mean differences in all muscles for each sex group. The mean differences were compared by examining their 95% CIs. The differences were regarded as significant when CIs did not overlap. Based on these differences, we established the patterns of relationship. These patterns were then used to compare sex differences.

In the final step, to study sex differences in the relationships in the muscles, we calculated mean ratios in the Vrn of muscle paired, and unpaired *t*-tests or Mann-Whitney *U* tests were used to compare those mean ratios between female and male individuals. Significance tests were 2-tailed, and α was set at 0.05.

Results

Sex Differences in Isolated Muscles

The unadjusted analyses showed that only the biceps femoris had a significant mean difference ($p = 0.003$; $d = 0.73$) between the 2 sex groups as shown in Table 1. The mean VC of the biceps femoris was 6.66 mm·s^{-1} lower in female individuals than in male individuals. However, when comparisons were made with the means adjusted by anthropometric corrections, only the rectus femoris presented differences ($p < 0.001$) between sexes. The rectus femoris in female individuals was 6.20 mm·s^{-1} slower than that in male individuals.

Sex Differences in Muscle Relationships

The CI comparisons between the hamstring and quadriceps within each sex group show that both female and male individuals have a similar pattern among the 5 muscles studied. These patterns are shown in Figure 2. On the one hand, the Vrn of the hamstring was significantly lower than that of the quadriceps in both sex groups (ratios ranging from 38.1 to 63.3%). Nevertheless, female individuals showed a more pronounced difference in the VC between biceps femoris and quadriceps (range: 18–26 mm·s^{-1}) than male individuals (range: 13–19 mm·s^{-1}). On the other hand, no statistical differences were found in each sex group in the muscle Vrn within the hamstring or the quadriceps muscular groups.

The biceps femoris/semitendinosus ratio in the hamstring was significantly higher ($p < 0.016$; $d = 0.70$) in male individuals than in female individuals (124.8 vs. 87.4%), as shown in Table 2. By contrast, among synergist pairs of the quadriceps muscles, no male-female differences were found and all ratios presented values close to 100%. Between agonist-antagonist muscle pairs from the hamstring and quadriceps, there were significant sex differences only in the relationship between biceps femoris and quadriceps. The biceps femoris/rectus femoris ($p < 0.045$; $d = 0.64$), biceps femoris/vastus lateralis ($p < 0.018$; $d = 0.72$), and biceps femoris/vastus medialis ($p = 0.015$; $d = 0.50$) ratios were significantly lower in female individuals than in male individuals (17.9, 18.8, and 15.0%, respectively), representing a medium effect size. Semitendinosus ratios with the quadriceps remained close to 50% in both sexes. Only the ratios of the biceps femoris in its relation with the other muscles showed sex differences greater than 15%, as shown in Figure 3.

Discussion

In this study, we examined whether there are sex differences among recreationally active young adults in the VC of both individual hamstring and quadriceps and also in the relationship between their VC ratios. Two important findings emerged: first, the rectus femoris in female individuals is slower than that in male individuals when data are adjusted by anthropometric variables, and second, both male and female individuals have a similar

Table 1

Vrn of the hamstring and quadriceps by sex and differences between sexes.*†

	Sex groups		Sex differences (female individuals minus male individuals)	
	Mean; 95% CI (*SD*)		Mean (95% CI); effect size ‡	
Muscle	Female individuals	Male individuals	Unadjusted	Adjusted §
Biceps femoris	14.41; 13.06–15.76 (3.93)	21.03; 17.04–25.01 (11.43)	−6.66 (−10.69 to −2.64); 0.73‖	−3.45 (−9.88 to 2.67)
Semitendinosus	16.70; 16.03–17.38 (1.97)	16.91; 15.99–17.84 (2.65)	−0.32 (−1.45 to 0.81); 0.14	−0.78 (−2.60 to 1.04)
Rec-F	32.03; 30.66–33.41 (4.01)	34.19; 32.30–36.08 (5.42)	−2.05 (−4.32 to 0.20); 0.42	−6.20 (−9.59 to −2.80)‖
Vastus lateralis	37.57; 36.06–39.08 (4.39)	37.81; 36.05–39.56 (5.02)	−0.17 (−2.40 to 2.06); 0.07	−2.98 (−6.43 to 0.47)
Vastus medialis	40.21; 37.37–43.06 (8.29)	40.01; 38.28–41.74 (4.96)	0.21 (−3.09 to 3.50); 0.03	−2.70 (−7.95 to 2.54)

*Vrn = normalized response velocity; CI = confidence interval.
†All values in mm·s^{-1}.
‡Effect size was estimated with Cohen's *d* only for the unadjusted model.
§Mean differences adjusted by body mass and height.
‖Significant differences at $p \leq 0.05$.

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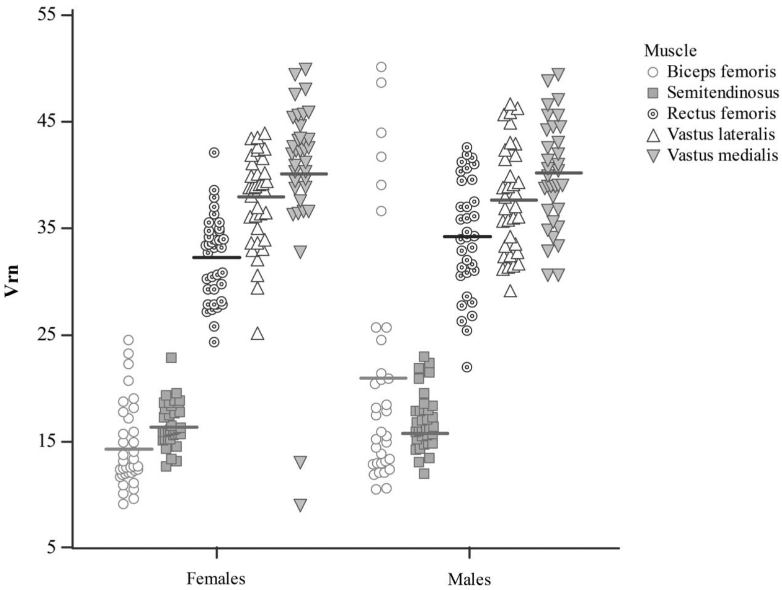

Figure 2. Scatterplot with means of the normalized response velocity (Vrn) of all muscles in each sex group.

pattern among the VC of the quadriceps and hamstring. Overall, the hamstring is slower than the quadriceps, but female individuals showed a more pronounced difference in the VC between biceps femoris and quadriceps than male individuals. As a consequence, the ratios between the VC of biceps femoris and quadriceps were lower in female individuals than in male individuals. By contrast, the ratios between the VC of semitendinosus and quadriceps were similar.

The unadjusted analysis showed that the biceps femoris was significantly slower in female individuals (6.66 $mm \cdot s^{-1}$ difference) than in male individuals. This finding is consistent with previous studies (29). However, there are no differences in the biceps femoris between female and male individuals when adjusted by anthropometric variables. This finding is plausible because in other mechanical contractile properties, such as stiffness (6) or strength (13), there are no differences in the hamstring between sexes when data are normalized by body mass and height.

Previous research has reported greater normalized strength (15) and normalized muscular activation during functional tasks (11) of the quadriceps in female individuals compared with male individuals. Because the VC in our study showed the opposite mechanical contractile properties, it became an unexpected finding. Specifically, we found that the differences in the VC of the quadriceps increased at least 100% of their unadjusted values after adjustment for body mass and height. Thus, our finding evidences the need to take into account the anthropometric characteristics when making comparisons between sexes. We believe that the potential reasons for this unexpected finding could be highlighted through studies that simultaneously measure strength, activation, and/or the VC in both sexes. Thus, the next investigations delving into this issue should compare these muscle properties and find out whether their behaviors are similar when their differences are adjusted by anthropometric data.

Table 2

Ratios‡ of the contraction velocity between the pairs of muscles.*

	Ratios (%) (95% CI)		Sex differences (female individuals minus male individuals)	
Pairs of muscles	Female individuals	Male individuals	Mean difference (95% CI)	Effect size
Between the hamstring and quadriceps				
Biceps femoris/rectus femoris	45.6 (41.1–50.2)	63.3 (50.8–75.8)	−17.9 (−30.6 to −5.27)†	0.64
Biceps femoris/vastus lateralis	38.1 (34.9–42.7)	57.4 (45.7–69.0)	−18.8 (−30.5 to −7.0)†	0.72
Biceps femoris/vastus medialis	40.0 (31.9–48.1)	55.1 (42.8–67.3)	−15.0 (−29.4 to −0.7)†	0.50
Semitendinosus/rectus femoris	52.8 (50.0–55.6)	50.6 (46.9–54.3)	1.7 (−2.9 to 6.3)	0.18
Semitendinosus/vastus lateralis	45.1 (42.4–47.8)	45.5 (42.4–48.3)	−0.7 (−4.6 to 3.2)	0.09
Semitendinosus/vastus medialis	47.3 (36.3–58.2)	42.9 (42.5–48.3)	4.3 (−6.9 to 15.6)	0.18
Within the hamstrings				
Biceps femoris/semitendinosus	87.4 (78.9–95.9)	124.8 (102.2–147.4)	−37.1 (−60.2 to −13.9)†	0.70
Within the quadriceps				
Rectus femoris/vastus lateralis	86.27 (81.3–91.2)	90.8 (86.5–95.1)	−4.5 (−10.8 to 1.8)	0.33
Rectus femoris/vastus medialis	88.3 (71.6–104.9)	86.3 (81.0–91.6)	1.9 (−15.4 to 19.3)	0.05
Vastus lateralis/vastus medialis	104.8 (82.9–126.7)	95.2 (90.9–99.5)	9.6 (−12.6 to 31.8)	0.21

*CI = confidence interval.
†Significant differences at $p < 0.05$.
‡All ratios are expressed in percent.

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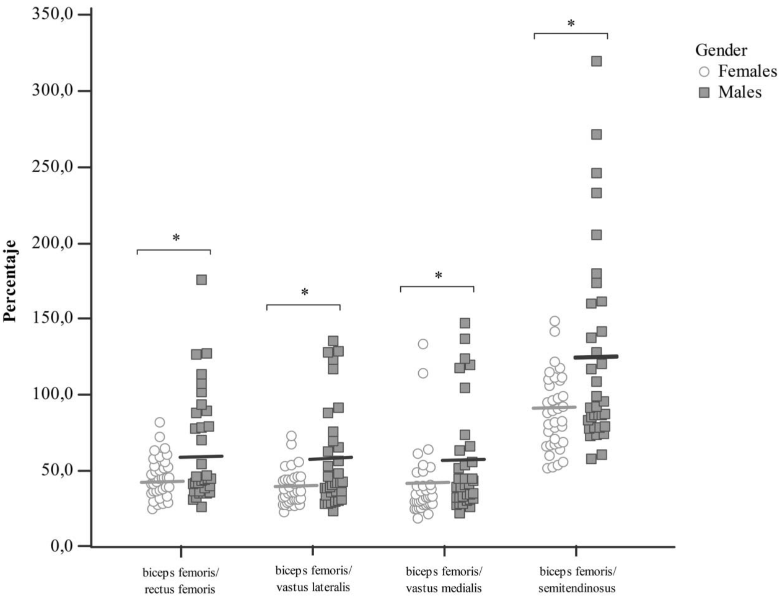

Figure 3. Ratios of the velocity of contraction between the pairs of muscles by sex (only pairs with differences higher than 15% are shown). *Significant differences at $p < 0.05$.

With respect to the relationship between the hamstring and quadriceps, both sexes showed VC global differences between both muscle groups, as it also happens with strength (16) and activation (11), with lower values for the hamstring. Therefore, our study supports that sex is a source of nonhomogeneous mechanical contractile properties between the hamstring and quadriceps. Nevertheless, this study specifically added to this debate that sex differences could also be different in the muscles. Thus, it is evidenced that the VC of the biceps femoris in female individuals, as opposed to the VC of the semitendinosus, showed a more pronounced difference with the quadriceps in comparison with male individuals. These sex differences between the biceps femoris and quadriceps ratios (around 20%) were similar to previously reported ratios for strength (16) and muscular activation (11). This study's authors regarded these differences as one of the influential factors in a greater risk of injury in female individuals. Given that our level of VC differences is similar to theirs, we also regard that the VC can be a relevant factor. Nevertheless, we consider that new research should be conducted to clarify the relationship between the VC and other similar constructs (e.g., muscle performance).

Our study also evidenced sex differences in the relationship between the semitendinosus and biceps femoris. Whereas the biceps femoris was slower than the semitendinosus in female individuals, the biceps femoris was faster in male individuals. This could indicate different performance percentages per sex in some of their functions, such as tibia control. This consideration could also be supported because the lower role in the VC of the biceps femoris for female individuals also occurs in relation to its role in other muscular aspects apart from the VC. In this regard, previous research had observed how the biceps femoris had less activity pattern with respect to its main action direction than the semitendinosus in female individuals compared with male individuals (22).

To our knowledge, no previous studies have calculated the VC ratio for the relationship between the hamstring and quadriceps, nor have they incorporated the semitendinosus in their sex comparisons. Furthermore, our study is the first to adjust sex differences in the VC by anthropometric data. Moreover, the use of a standardized measuring technique and objective measurements that allow for comparison with other studies could be considered a strength of the study.

Our study had several limitations. First, according to previous studies (29), we adjusted the differences between sexes by anthropometric measures but did not take into account other measures, such as adiposity. In addition, adjustments for morphologic properties of the muscle (e.g., cross-sectional area) could be more appropriate. Second, we used a technique that measures the VC by submaximal stimulated muscle activation and this could limit inferences to performance capabilities and injury risk. Nevertheless, it has been previously used by other authors (30) who have reported it as a possible cause of joint instability. In addition, being the first time that the VC ratios were obtained using TMG, the mechanical contractile properties recorded are limited to the characteristics of our sample. Therefore, it is unknown whether the findings would be similar in other populations or whether this ratio has validity as a predictor of the performance or the risk of injury.

Practical Applications

Our study provides several research and clinical implications. First, because the hamstrings and quadriceps showed imbalances in both sexes (especially the biceps femoris with the quadriceps in female individuals), these imbalances could be an objective of interventions when attempting to reduce the differences between the muscle groups. To monitor the changes produced by these interventions, TMG can serve as a tool for evaluation. Second, our study provides the VC ratios as a new possibility to analyze the relationship at the muscle level. Furthermore, as our ratios were established in healthy young people, they can be used for comparison with other nonhealthy populations.

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ARTÍCULO III

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PeerJ

Tensiomyographical responsiveness to peripheral fatigue in quadriceps femoris

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ABSTRACT

Background. Fatigue influences athletic performance and can also increase the risk of injury in sports, and most of the methods to evaluate it require an additional voluntary effort. Tensiomyography (TMG), which uses electrical stimulation and a displacement sensor to evaluate muscle contraction properties of one or more muscle bellies, has emerged as a technique that can assess the presence of peripheral and central fatigue without requiring additional voluntary efforts. However, the evaluation of the TMG's ability to detect fatigue is limited, both at the level of muscle bellies and statistical methods. Thus, the aim of the present study was twofold: (i) to examine and compare the tensiomyographical responsiveness to quadriceps femoris (QF) fatigue by multiple statistical methods and (ii) to analyze sex differences in the variation produced by fatigue in TMG parameters.

Methods. Thirty-nine recreational athletes participated (19 males/20 females; aged 22 ± 2 years). TMG parameters of QF bellies and maximal voluntary isometric contraction (MVIC) were measured before and after a fatigue protocol. TMG parameters used were maximum radial deformation (Dm), contraction time between 10–90% of the Dm (Tc), contraction velocity between 10–90% (Vc) and of the first 10% (V10) of the Dm. Internal responsiveness of TMG to fatigue was analyzed by paired t-test and standardized response mean (SRM). External responsiveness was examined by correlations, regression models, and receiver operating characteristic (ROC) curves.

Results. All TMG parameters, except for Tc of rectus femoris and vastus medialis, showed large internal responsiveness. In adjusted regression models by sex, only Dm and V10 of rectus femoris were statistically associated ($p < 0.05$) with b coefficients of 0.40 and 0.43, respectively. r2 explained the 22% of the total variance. In addition, these parameters could discriminate between QF with and without fatigue.

Conclusion. Since the QF is the main strength contributor during multiple physical activities, clinicians and trainers will be able to discriminate the presence of fatigue and the magnitude of changes in the QF strength by TMG evaluation.

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Keywords Responsiveness, Tensiomyography, Fatigue, Quadriceps, Sex, Recreational athletes

INTRODUCTION

Fatigue is defined as a decline in muscular performance which produces a reduction in strength and power generation (*Ditroilo et al., 2011*). It can be further explained by

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factors related to the central nervous system as changes at the spinal level (*Gandevia, 2001*) or by peripheral factors associated to the muscle, such as failure of transmission at the neuromuscular junction (*Allen, Lamb & Westerblad, 2008*). Its manifestation can vary in subjects with different training backgrounds (*Garrandes et al., 2007*), type of muscle contraction performed (*Kay et al., 2000*), or even between sex (*Albert et al., 2006*; *Martin & Rattey, 2007*; *Ansdell et al., 2017*).

Since fatigue influences athletic performance (*Thorlund et al., 2008*; *Ditroilo et al., 2011*) and can also increase the risk of injury in sports (*Zebis et al., 2011*; *Liederbach et al., 2014*), its study has been of interest. Multiple methods have been used to induce fatigue, both central fatigue in several muscle groups or peripheral fatigue in a specific muscle (*García-Manso et al., 2011*; *Hunter et al., 2012*; *Macgregor et al., 2016*; *Wiewelhove et al., 2017*; *Wiewelhove et al., 2018*). Thus, fatigue has been evaluated after short term (*Macgregor et al., 2016*; *Abelairas-Gómez et al., 2018*) and long duration efforts, such as several days of intense training sessions (*Wiewelhove et al., 2017*), and also after isolated long sessions (2–12 h approximately) (*Lepers et al., 2002*; *García-Manso et al., 2011*; *Wiewelhove et al., 2018*).

The most used fatigue evaluation methods have been based on changes in maximal voluntary isometric contractions (MVICs) (*Lepers et al., 2002*; *Zebis et al., 2011*) , muscle activation (*Garrandes et al., 2007*; *Thorlund et al., 2008*), kinematics and kinetics measurements (*Liederbach et al., 2014*; *Tam et al., 2017*), biochemical markers (*Gorostiaga et al., 2012*), or muscular contractile properties (*García-Manso et al., 2011*; *De Paula Simola et al., 2016*). In a situation of fatigue, most of these methods would require an additional voluntary effort. Their application therefore would not be practical or safe facing the possible presence of central inhibition (*Graven-Nielsen et al., 2002*), or the possibility of increase any extant muscular damage (*Macgregor et al., 2016*).

Tensiomyography (TMG), which uses electrical stimulation and a displacement sensor to evaluate muscle contraction properties of one or more muscle bellies (*Valencic & Knez, 1997*), has emerged as a technique that can assess the presence of peripheral and central fatigue without requiring additional voluntary efforts (*García-Manso et al., 2011*; *De Paula Simola et al., 2016*). Peripheral fatigue has been evaluated by TMG for specific muscle group from both lower and upper limbs (*Carrasco et al., 2011*; *Hunter et al., 2012*; *García-Manso et al., 2012*; *Macgregor et al., 2016*). In contrast, central fatigue has been evaluated only in the lower limb, being quadriceps femoris (QF) the most studied muscle group (*García-Manso et al., 2011*; *De Paula Simola et al., 2015*; *De Paula Simola et al., 2016*; *Giovanelli et al., 2016*; *Raeder et al., 2016*; *Wiewelhove et al., 2017*).

Responsiveness is defined as the ability of a tool to detect important clinical changes over time (*Guyatt et al., 1989*). Since this characteristic is essential to assess fatigue by TMG, it has been analyzed by multiple studies (*García-Manso et al., 2011*; *Hunter et al., 2012*; *De Paula Simola et al., 2015*; *De Paula Simola et al., 2016*; *Giovanelli et al., 2016*; *Macgregor et al., 2016*; *Raeder et al., 2016*; *Wiewelhove et al., 2017*; *Abelairas-Gómez et al., 2018*). Most of these studies evaluated one muscle belly and they used one or two statistical methods of either internal responsiveness (e.g., paired t-test and effect size) or external responsiveness (correlation with reference measure or regression models) Internal responsiveness is the ability of a measure to change over a set period and external responsiveness reflects the extent to which changes in a measure over a specified time frame related to corresponding changes in an external reference measure of health status (*Husted et al., 2000*). Overall, TMG of those evaluated muscle bellies has shown to be internally and externally responsive in assessing central fatigue (*García-Manso et al., 2011*; *De Paula Simola et al., 2015*; *De Paula Simola et al., 2016*; *Giovanelli et al., 2016*; *Raeder et al., 2016*; *Wiewelhove et al., 2017*), and internally responsive to peripheral fatigue (*Hunter et al., 2012*; *García-Manso et al., 2012*; *Macgregor et al., 2016*; *Abelairas-Gómez et al., 2018*). However, to the best of our knowledge, the external responsiveness of TMG has not been yet assessed for peripheral fatigue, and therefore comparisons between internal and external responsiveness has not been established. Furthermore, to our knowledge, TMG responsiveness has not been simultaneously evaluated in multiple bellies, neither analyzed by by multiple statistical indicators of responsiveness. At the same time, understanding the mechanisms behind the changes in TMG parameters caused by fatigue in both sexes, is also an area of research that needs further development.

Therefore, the primary objective of our study was to examine and compare the responsiveness of TMG parameters to QF peripheral fatigue of three muscle bellies (rectus femoris (RF), vastus lateralis (VL), and vastus medialis (VM)) by multiple statistical methods. A secondary objective was to examine whether there are differences between sex in the variation produced by fatigue in TMG parameters. Our hypotheses were: QF bellies have different responsiveness to peripheral fatigue; and the changes of TMG parameters are similar between males and females.

MATERIALS & METHODS

Study design

A single group pretest-posttest design was used, which involved repeated TMG and MVIC measures of the dominant lower limb QF before and after a fatigue protocol within the same session. Participants were physiotherapy students recruited by email using the University of Valencia Intranet. This study was conducted from April to July 2018. All measurements were carried out between 10 a.m. and 2 p.m in the clinical research laboratory of the Department of Physiotherapy (University of Valencia) at an ambient temperature 21–22 °C. An experienced examiner in the measurement techniques evaluated the participants. He was a physiotherapist who had used TMG and hand dynamometers both in research and in clinical practice for several years. Before participation, participants were informed of the study procedures and their possible associated risks. All of them provided written informed consent. This study was completed following the principles outlined in the Declaration of Helsinki and it was approved by the Ethics Committee of the University of Valencia (Spain) (H1523633864087).

Participants

Thirty-nine recreational athletes were evaluated. All participants performed exercise 3 times per week and practiced activities such as running, swimming, cycling, or central strength training. The specific inclusion criteria were: (a) aged between 18 and 30 years, (b) not surgically

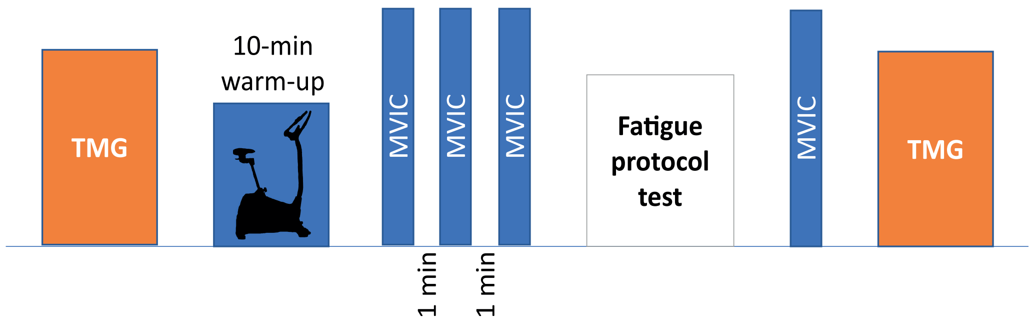

Figure 1 **Schematic representation of experimental procedures.** TMG, tensiomyography; MVIC, maximal voluntary isometric contraction.

Full-size DOI: 10.7717/peerj.8674/fig-1

operated on the lower limb, (c) without pain in the lower limb in the 2 months before data collection, and (d) performing physical exercise a minimum of 3 days per week. The exclusion criteria were: (a) practicing a specific sport as an amateur or professional, (b) contraindication to the use of electrodes due to injury or allergy to the adhesive, and (c) nontolerance to electrical stimulation.

Procedures

Before starting the session, height was measured using a 1-millimeter sensitivity flexible tape measure, while body mass and body mass index (BMI) were assessed using a standardized body composition analyzer (Tanita BC 418 MA, Tanita Corp, Tokyo, Japan). Next, TMG parameters were measured and then, participants performed a warm-up, which consisted of 10 min cycling at comfortable speed (80 revolutions per minute) with low resistance and the performance of three submaximal isometric contractions of isometric knee extension (*Martins et al., 2017*). Following this, the MVIC test was performed. After the fatigue protocol, the order of the tests was reversed, and the strength test was performed first to reduce the time between MVIC and TMG tests in acute fatigue. A schematic representation of the experimental procedures is reported in the Fig. 1.

Tensiomyography measurements

First, participants were placed supine and resting on the stretcher. The knee was placed at 120° of flexion (considering full extension at 180°), fixing such position with a triangular foam cushion (*García-García et al., 2016*; *Martín-San Agustín et al., 2020*). The area where the TMG sensor and electrodes were placed was shaved and cleaned with gauze and alcohol. The position of the sensor for each QF belly was determined using the anatomical criteria described in the literature (*Dahmane et al., 2005*; *Tous-Fajardo et al., 2010*; *Rey, Lago-Peñas & Lago-Ballesteros, 2012*). This position was marked with a permanent marker so that it would remain throughout the evaluation. The sensor was finally placed on this point perpendicularly to the thigh and the electrodes were placed at five cm distance from it, forming an imaginary straight line along the belly (Fig. 2).

The contractile properties of each belly were evaluated during an maximal elicited contractions with the TMG electro stimulator (TMG-100 System). Starting from 20 mA

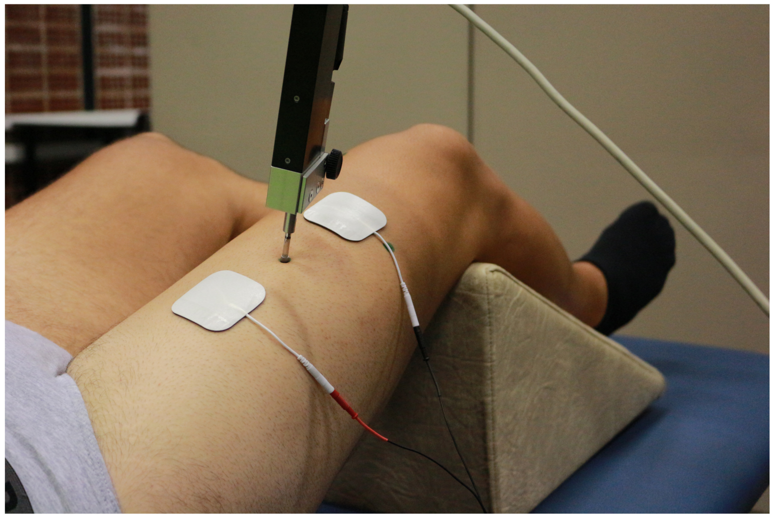

Figure 2 **Tensiomyographical measurement of rectus femoris.** Photo credit: Rodrigo Martín-San Agustín.

Full-size DOI: 10.7717/peerj.8674/fig-2

with 1ms pulses, each stimulation was increased by 10mA until achieving the maximum radial deformation (Dm) of the muscular belly. A time of 10s was left between stimuli to minimize fatigue or potentiation effects (*Krizaj, Simunic & Zagar, 2008*). Before data acquisition, a pilot test was done to verify the functioning of the TMG. For each belly, spatial and temporal parameters were measured: Dm, contraction time between 10 and 90% of the Dm (Tc), contraction velocity between 10 and 90% of the Dm (Vc), and contraction velocity of the first 10% of the Dm (V10). TMG has proven to be a method with a high relative [ICC for Dm (0.91–0.99), Tc (0.70–0.98), and VC > 0.95] and absolute (low coefficient of variations for Dm, Tc, and VC) reliability (*Martín-Rodríguez et al., 2017*; *Lohr et al., 2018*).

Maximal voluntary isometric contraction test

MVIC of the QF was measured by a MicroFET2 handheld dynamometer (Hoggan Health Technologies Inc., Salt Lake City, UT). Participants were seated in an isokinetic dynamometer (Prima Plus, Easytech, Italy) with their torso and hips tied so they were stable, and with a 90° hip flexion. MVIC was evaluated in 90° knee flexion, considering 0° the complete extension (Fig. 3). MicroFET2 was fixed with a rigid belt perpendicular to the ankle five cm above the malleoli, with a pad between the tibia and the dynamometer to minimize the discomfort caused by the contact (*Hansen et al., 2015*).

After the warm-up, participants completed three MVIC for 5s, with a 60-second rest after each repetition. Through verbal stimuli, participants were instructed to exert and maintain the maximum effort during the session. MicroFET2 has proven to be a valid method to measure the MVIC of the QF with an excellent inter-examiner reliability (ICC: 0.93, 95% CI [0.83–0.97]) and a minimal detectable change (MDC) of 14.1 N*m (95% CI [9.23–22.01]) (*Hansen et al., 2015*).

Figure 3 **Maximal voluntary isometric contraction test for quadriceps femoris.** Photo credit: Rodrigo Martín-San Agustín.

Full-size DOI: 10.7717/peerj.8674/fig-3

Fatigue protocol test

After performing the baseline measurements, participants were requested to implement a protocol based on a 60s fatiguing isometric contraction at 70% MVC (*Melchiorri & Rainoldi, 2011*). The experimental setup was the same as the one adopted during the MVIC test. The handheld dynamometer, previously set at 70% MVIC, was used to display the feedback (*Melchiorri & Rainoldi, 2011*). It was considered that the fatigue was achieved when the reduction of the MVIC was higher than the upper limit of the MDC reported in a previous study (22.01 N*m) (*Hansen et al., 2015*).

Statistical analysis

Baseline data were summarized as means and standard deviations (SD) for continuous variables and as absolute and relative frequencies for categorical variables. Variables were checked for normality with the Kolmogorov–Smirnov test and homogeneity of variances with Levene's test. A summary was also provided for participants with and without fatigued QF.

Paired t-tests were used to compare changes in the TMG parameters and MVIC within each sex group. These changes were also compared between sex groups by using non-paired t-tests.

Internal responsiveness was determined by the paired t-test and supplemented with an effect size statistic, as recommended by *Husted et al. (2000)* [30]. To reduce the probability of getting false positives, we increased the acceptance level from 0.05 to 0.01 for paired t-test because multiple comparisons were made on the same data set. Of the current effect size statistics we used the standardized response mean (SRM), which provides an estimate of the magnitude of change that is not influenced by sample size (*Navarro-Pujalte et al., 2018*). It was calculated as (MeanFollowup _ MeanBaseline)/Standard deviationFollowup-Baseline and the 95% confidence intervals were calculated using the bootstrapping estimation method. Values of 0.20, 0.50, and 0.80 or higher have been proposed in the literature (*Husted et al., 2000*) to represent small, moderate, and large responsiveness, respectively. Besides, we calculated the percentage of participants that exceeded MDC. This statistic examines the extent to which change score exceeds the amount of variability accounted by measurement error (*Pardasaney et al., 2012*), which is calculated as $SEMx1.96x\sqrt{2}$, where SEM is the standard error of measurement.

External responsiveness was determined by correlations, regression models, and receiver operating characteristic (ROC) curves (*Husted et al., 2000*). The external criterion for assessing the external responsiveness of the TMG tool was the magnitude of change in the MVIC.

We assumed that: (i) changes in the external criterion (MVIC) in participants with fatigue would be associated with changes in the TMG parameters; (ii) participants without fatigue would have the smallest change in the TMG parameters (and therefore change in these TMG parameters can be useful to classify participants' QF as fatigued or not fatigued). To test the first hypothesis, correlations and simple and multiple linear regression models were used. In the regression models the explanatory variable was the change of each TMG parameter while the response variable was the change in MVIC between before and after protocols. Each model was controlled by sex, and comparisons were carried out between the presence or absence of this control. Goodness-of-fit of the model was assessed by r2. To test the second hypothesis, we calculated the area under the ROC curve (AUC), which represents the probability that the measure of correctly classifying participants has (*Husted et al., 2000*). An AUC >0.70 was used as a generic benchmark to consider acceptable its discriminant ability (*Menaspà, Sassi & Impellizzeri, 2010*).

For sample size calculation, we selected the multiple regression as the main statistic of responsiveness because it allowed us to examine change relationships controlling by a covariate relevant in our study (sex). Regarding this statistic, we used the usual rule of thumb that 15 participants per predictor are needed for a reliable equation in multiple regression models (*Tabachnick & Fidell, 2007*). We recruited a minimum of 30 participants assuming a maximum of 2 explanatory variables (TMG parameter and sex). Statistical significance was set at $p < 0.05$. All analyses were performed using the Statistical Package for the Social Sciences software program (SPSS version 24.0; IBM SPSS, Chicago, IL, USA).

RESULTS

Participants' characteristics

Baseline characteristics of participants are listed in Table 1. A total of 35 (89.7%) participants achieved QF fatigue after the application of the fatigue protocol. They were 19 of 20 females (95%) and 16 of 19 males (84.2%). Participants with and without fatigue showed no significant differences ($p > 0.05$) in any of their baseline characteristics.

Changes associated with the fatigue protocol

Participants with peripheral fatigue ($n = 35$) had a significant decrease (31.5%) on their MVIC after the fatigue protocol (from 203.3 N*m to 138.9 N*m). Table 2 shows that both sex groups had a similar pattern of change: males reduced 30.8% and females 32.1%. Table 2 also shows patterns of change by sex groups for TMG parameters of the RF, VL, and VM. All these parameters, except for the Tc of the RF and VM, had significant differences within but not between sex groups.

Figure 4 shows changes in TMG parameters for all participants with peripheral fatigue. All parameters, except for Tc, showed a significant difference ($p < 0.001$) for the three bellies of the QF. Dm's decrease ranged from 18.22% to 21.65%; Vc decreased from 15.62 to 22.20%, and V10 decreased from 14.80% to 23.77%.

Internal and external responsiveness

Internal and external TMG responsiveness to fatigue of QF bellies is shown in Table 3. Internal responsiveness statistics suggest that all TMG parameters, except for Tc of RF and VM, showed large internal responsiveness (SRM > 0.8) among participants with QF fatigue. Dm and V10 in RF were the parameters in which most of the participants exceeded the MCD (91.3% and 97.1%, respectively). Only Dm, Vc, and V10 of the RF showed to be linearly associated with changes in the MVIC. After controlling by sex, adjusted models typically provided b coefficients and r2 with small variations regarding their respective unadjusted model (range 0.01 to 0.05). Consequently, Dm and V10 of RF were still statistically associated with b coefficients of 0.40 and 0.43, respectively. Moreover, the models of these parameters explained the 22% of the total variance.

The AUC analysis suggests that changes of several TM G parameters (Dm in RF and VL, Tc in VL, and V10 in RF and VM) were >0.70 and could discriminate between QF with and without fatigue. Also, the overlapping among their 95% CI suggests that none of these TMG parameters is superior to the others to discriminate fatigue.

Table 1 **Baseline characteristics of the participants in total and separated by fatigued condition.**

Baseline Characteristics	Total ($n = 39$)	Fatigued participants ($n = 35$)	Non-fatigued participants ($n = 4$)
Males/females, N (%)	19 (48.7%)/20 (51.3%)	16 (45.7%)/19 (54.3%)	3 (75%)/1 (25%)
Age (years)	22 (2)	22 (2)	21 (1)
Physical activity (minutes)	316.5 (180.8)	314.6 (186.7)	332.5 (136.9)
Anthropometric			
Body mass (kg)	67.37 (13.42)	66.10 (11.12)	78.55 (12.05)
Stature (cm)	173.3 (9.50)	172.5 (9.09)	180.7 (11.24)
BMI (kg/m2)	22.22 (2.72)	22.02 (2.71)	24 (2.53)
QF strength			
MVIC (N*m)	207.56 (74.19)	203.31 (75.82)	244.72 (50.24)
Tensiomyography parameters			
Rectus femoris			
Dm (mm)	10.26 (1.42)	10.32 (1.44)	9.76 (1.28)
Tc (ms)	25.45 (4.04)	25.69 (3.95)	23.39 (4.84)
Vc (mm/s)	327.96 (58.59)	326.62 (69.76)	339.70 (53.04)
V10 (mm/s)	43.07 (5.32)	43.08 (5.39)	42.93 (5.33)
Vastus lateralis			
Dm (mm)	5.74 (1.11)	5.63 (0.94)	6.64 (2.04)
Tc (ms)	21.37 (3.02)	21.54 (3.11)	19.87 (1.35)
Vc (mm/s)	217.78 (50.10)	211.58 (39.81)	271.95 (97.28)
V10 (mm/s)	25.31 (5.18)	24.73 (4.21)	30.46 (9.98)
Vastus medialis			
Dm (mm)	4.57 (0.85)	4.52 (0.64)	5.08 (2.01)
Tc (ms)	19.60 (1.82)	19.61 (1.90)	19.48 (1.04)
Vc (mm/s)	187.22 (33.12)	185.08 (26.57)	205.93 (73.31)
V10 (mm/s)	23.22 (4.03)	22.97 (2.89)	25.37 (10.19)

Notes.

Date represents mean and standard deviation unless otherwise noted.

BMI, body mass index; Dm, maximal radial displacement; Tc, contraction time; Vc, contraction velocity between 10–90% of the Dm; V10, contraction velocity of the first 10% of the Dm; QF, quadriceps femoris; MVIC, maximal voluntary isometric contraction.

DISCUSSION

To our knowledge, this is the first study to evaluate the internal and external TMG responsiveness across a variety of QF muscle bellies to changes induced by peripheral fatigue. We found that TMG parameters Dm and V10 of the RF showed both internal and external responsiveness.

In our study, multiple statistical methods to evaluate the internal responsiveness (paired t-test and SRM) and external responsiveness (correlations, regression models and ROC) of the TMG were used, which is line with the recommendations of *Husted et al. (2000)*. In previous studies, most of these statistics have been used to evaluate only the TMG ability of change to fatigue (*García-Manso et al., 2011*; *De Paula Simola et al., 2015*). Thus, to the best of our knowledge, this is the first study to use several statistical methods to assess internal and external responsiveness. Furthermore, since most of the previous studies assessing fatigue by TMG have only evaluated isolated muscle bellies (*García-Manso et al., 2011*;

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Table 2 Differences within and between sex groups in the TMG parameters and MVIC after fatigue protocol.

Muscle	Males				Females			
	Baseline	Fatigued	Differences		Baseline	Fatigued	Differences	
			Mean (SD); *p*	%			Mean (SD); *p*	%
QF strength								
MVIC (N*m)	272.1 (51.0)	187.3 (40.1)	84.7 (37.8); <0.001	30.8	145.4 (30.7)	98.1 (24.4)	47.3 (22.3); <0.001	32.1
Rectus femoris								
Dm (mm)	9.91 (1.66)	7.46 (1.87)	2.45 (1.27); <0.001	25.2	10.67 (1.16)	8.71 (1.76)	1.95 (1.13); <0.001	18.7
Tc (ms)	24.58 (4.25)	24.52 (6.37)	0.06 (3.28); 0.941	1.1	26.62 (3.52)	27.63 (5.43)	−1.01 (4.42); 0.334	4.1
Vc (mm/s)	330.01 (78.95)	250.71 (66.81)	79.30 (48.65); <0.001	21.8	373.76 (39.15)	256.21 (51.02)	67.55 (42.26); <0.001	20.9
V10 (mm/s)	43.17 (6.55)	32.78 (7.72)	10.39 (5.35); <0.001	24.4	43.01 (4.37)	33.01 (5.13)	10.00 (4.20); <0.001	23.2
Vastus lateralis								
Dm (mm)	5.47 (1.18)	4.48 (0.76)	0.99 (1.10); 0.003	20.5	5.78 (0.70)	4.10 (1.15)	1.68 (0.90); <0.001	29.5
Tc (ms)	21.69 (3.05)	19.93 (4.31)	1.76 (2.44); 0.011	8.6	21.42 (3.24)	19.04 (1.88)	2.38 (2.15); <0.001	10.4
Vc (mm/s)	203.67 (49.77)	179.33 (66.24)	24.35 (43.77); 0.042	12.8	218.24 (28.76)	170.24 (37.41)	48.00 (43.15); <0.001	20.9
V10 (mm/s)	24.28 (5.04)	20.46 (6.78)	3.82 (4.33); 0.003	17.3	25.10 (3.45)	18.65 (4.66)	6.45 (4.55); <0.001	25.3
Vastus medialis								
Dm (mm)	4.69 (3.91)	3.91 (0.78)	0.78 (0.59); <0.001	16.3	4.37 (0.50)	3.51 (0.69)	0.86 (0.53); <0.001	19.8
Tc (ms)	20.25 (1.78)	19.96 (2.66)	0.28 (1.97); 0.573	1.4	19.07 (1.88)	18.26 (1.88)	0.81 (1.64); 0.045	3.9
Vc (mm/s)	186.06 (30.93)	159.90 (25.72)	29.16 (22.46); <0.001	14.9	184.26 (23.12)	153.76 (29.26)	30.50 (26.86); <0.001	16.2
V10 (mm/s)	23.76 (3.19)	21.09 (3.95)	2.67 (2.97); 0.003	11.2	22.31 (2.51)	18.33 (3.40)	3.98 (2.74); <0.001	17.8

Notes.

SD, standard deviation; Dm, maximal radial displacement; Tc, contraction time; Vc, contraction velocity between 10–90% of the Dm; V10, contraction velocity of the first 10% of the Dm; QF, quadriceps femoris; MVIC, maximal voluntary isometric contraction.

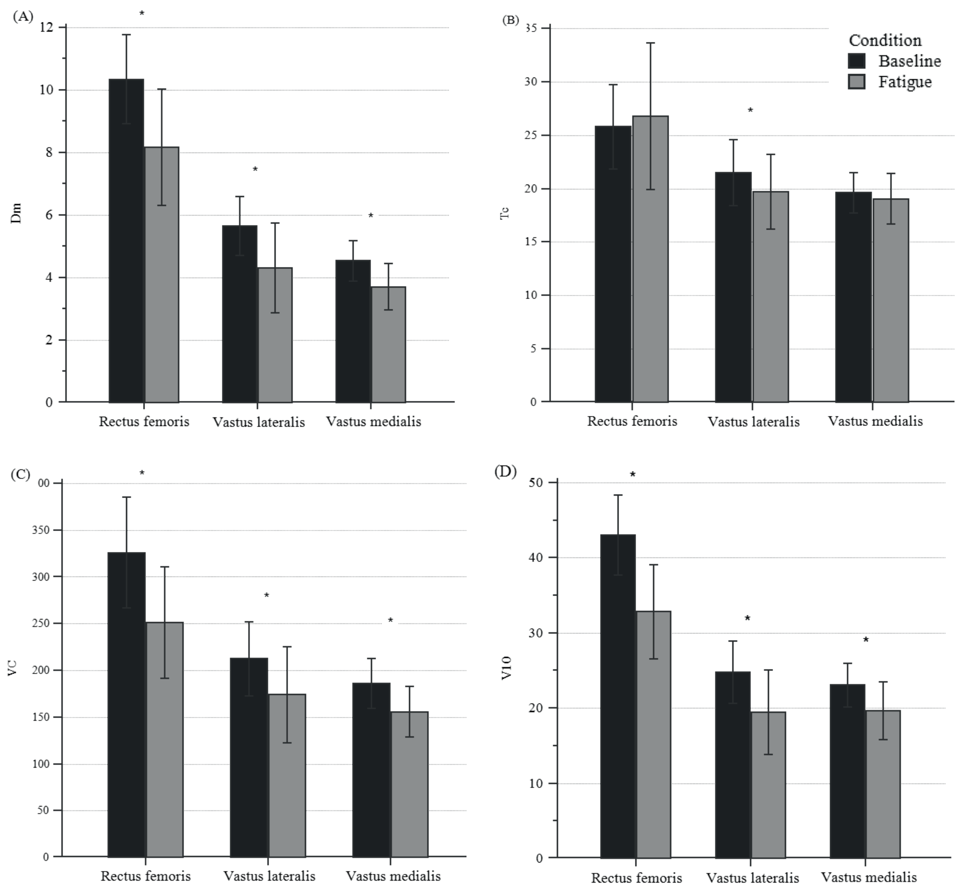

Figure 4 Differences in TMG parameters of quadriceps bellies between pre- and post-fatigue in all participants. (A) Differences in Dm, (B) in Tc, (C) in VC, and (D) in V10. *Significant differences set at $p < 0.01$; Specific p-values are shown in Table 3.

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Hunter et al., 2012; *De Paula Simola et al., 2015*; *De Paula Simola et al., 2016*; *Giovanelli et al., 2016*; *Macgregor et al., 2016*; *Raeder et al., 2016*; *Wiewelhove et al., 2017*), our study presents novel findings in the evaluation of TMG across multiple muscle bellies.

Regarding the internal responsiveness, large and negative SRM of the TMG parameters were found in most of the muscle bellies. Overall, our results are consistent with previous studies that induced peripheral and central QF fatigue (i.e., selective QF fatigue or caused in the entire lower limb musculature). Therefore, the reduction of RF TMG parameters is consistent with previous studies using peripheral (*Carrasco et al., 2011*) or central fatigue (*De Paula Simola et al., 2015*), finding them reductions in Dm, VC, or V10 after fatigue due to cycling or strengthening. On the other hand, the changes in VL and VM are also consistent with studies using central fatigue caused by strengthening programs (*De Paula Simola et al., 2016*; *Raeder et al., 2016*). In addition, Dm results showed consistence with other studies that induced peripheral fatigue in muscles such as the biceps brachii (*Hunter et al., 2012*; *García-Manso et al., 2012*) or the gastrocnemius medialis (*Macgregor et al., 2016*). These findings could be explained by changes in the pH (*Hunter et al., 2009*) and

Table 3 Responsiveness statistics for the TMG parameters.

Muscle	Internal responsiveness		% MCD	External responsiveness			
	Paired *t*-test (*p*)	SRM (95% CI)		Correlation method (Pearson's r and 95% CI); *p*	Linear regression method[a]		AUC (95% CI)
					b(SE); *p*	r2	
Rectus femoris							
Dm (mm)	0.001	−1.83 (−2.31; −1.47)	91.3	0.42 (0.12; 0.65); 0.004	0.40 (0.14); 0.007	0.22	0.73 (0.57; 0.86)
Tc (ms)	0.439	0.13 (−0.24; 0.39)	15.9	0.10 (−0.22; 0.40); 0.276	0.14 (0.15); 0.363	0.06	0.62 (0.45; 0.77)
Vc (mm/s)	0.001	−1.65 (−1.98; −1.30)	79.7	0.33 (0.02; 0.58); 0.020	0.26 (0.13); 0.052	0.13	0.59 (0.42; 0.74)
V10 (mm/s)	0.001	−2.20 (−2.65; −1.78)	97.1	0.45 (0.15; 0.67); 0.002	0.43 (0.15); 0.006	0.22	0.73 (0.57; 0.86)
Vastus lateralis							
Dm (mm)	0.001	−1.33 (−1.74; −0.82)	79.7	0.18 (−0.14; 0.47); 0.133	0.10 (0.12); 0.403	0.05	0.81 (0.65; 0.92)
Tc (ms)	0.001	−0.87 (−1.27; −0.41)	65.2	0.12 (−0.12; 0.48); 0.111	0.23 (0.19); 0.238	0.07	0.92 (0.79; 0.98)
Vc (mm/s)	0.001	−0.86 (−1.21; −0.46)	43.5	0.09 (−0.23; 0.39); 0.298	0.03 (0.11); 0.782	0.04	0.55 (0.39; 0.71)
V10 (mm/s)	0.001	−1.17 (−1.56; −0.71)	68.1	0.12 (−0.20; 0.42); 0.224	0.06 (0.12); 0.638	0.04	0.67 (0.50; 0.81)
Vastus medialis							
Dm (mm)	0.001	−1.46 (−1.84; −1.07)	76.8	0.12 (−0.21; 0.42); 0.116	0.09 (0.20); 0.643	0.04	0.65 (0.48; 0.79)
Tc (ms)	0.069	−0.34 (−0.72; 0.02)	42	−0.14 (−0.43; 0.18); 0.200	−0.28 (0.28); 0.331	0.06	0.52 (0.36; 0.68)
Vc (mm/s)	0.001	−1.17 (−1.50; −0.79)	68.1	0.17 (−0.15; 0.46); 0.143	0.17 (0.19); 0.364	0.06	0.68 (0.52; 0.82)
V10 (mm/s)	0.001	−1.14 (−1.47; −0.76)	71	0.26 (−0.06; 0.53); 0.054	0.25 (0.19); 0.194	0.08	0.76 (0.60; 0.88)

Notes.

SRM, standardized response mean; CI, confidence interval; MCD, minimal detectable change; SE, standard error; AUC, area under curve; Dm, maximal radial displacement; Tc, contraction time; Vc, contraction velocity between 10–90% of the Dm; V10, contraction velocity of the first 10% of the Dm.

[a]Adjusted by sex.

in different cellular molecules (e.g., Na+ or K+) (*Brody et al., 1991*), which cause damage in the sarcolemma and the reduction of the electrical stimulus, with a possible decrease in muscle displacement.

This study showed that Dm and V10 of RF had an acceptable external responsiveness in relation to our external criterion, namely changes in the strength evidenced by MVIC. As reflected by the regression coefficients, there was a moderate relationship between the amount of change in TMG parameters and strength scores. This relationship is consistent with a previous study using central fatigue (*De Paula Simola et al., 2015*). Furthermore, Dm and V10 were relevant according to sex, which can be explained by the fact that our sample showed similar change magnitudes in both TMG parameters and strength scores.

The fatigue protocol used in this study was highly effective (most of the QF showed fatigue). Males and females had similar strength change scores (Table 2). Previous studies reported different strength change scores between sexes when intensities between 25–50% of MVIC were used (*Clark et al., 2005*; *Ansdell et al., 2017*). In our study, an intensity of 70% of MVIC was used, suggesting that as the contraction intensity increase, the sex differences in muscle fatigue decrease , (*Hunter, 2014*). Therefore, future investigations should examine whether sex differences in strength changes are detected by sex differences in the TMG changes.

Our present study also showed that TMG has discriminative ability to classify the participants' QF as having fatigue or not after the application of the protocol. Dm and V10 of the RF also were two of the four parameters with this discriminative ability. This finding is partially consistent with previous studies (*Wiewelhove et al., 2017*), who examined AUC of RF after central fatigue in elite young athletes. Nevertheless, while AUC values of V10 shown in this study was similar to their results, AUC values of Dm was higher than previously published (*Wiewelhove et al., 2017*). Differences may be explained by the different type of fatigue (central fatigue caused by several training sessions of high-intensity interval training vs peripheral fatigue by an MVIC test) or by the athletes' training background (junior tennis players vs recreational athletes). Other parameters with that discriminative ability were Dm and Tc of VL, and V10 of VM. Since this ability was not previously analyzed in these muscle bellies (VL and VM), results of the actual study supplements earlier findingswhich have only evaluated AUC for external responsiveness of the TMG in RF (*Wiewelhove et al., 2017*) and it provides evidence to expand the application of the TMG to discriminate fatigue.

Actual study has several limitations. First, we used a fatigue protocol based on MVIC, which induces peripheral fatigue. Therefore, our findings would be limited to be extrapolated to others fatigue situations (e.g., concentric contractions). Second, our study was conducted with recreational athletes (i.e., anyone participating in an aerobic or athletic activity at least three times per week) (*Heinert et al., 2008*). Since the contractile properties of the muscle are conditioned by the type of exercise performed (*Loturco et al., 2015*), future research should compare our results with findings from athletes of different sports.

Our study found that most of the TMG parameters showed an acceptable internal responsiveness of QF peripheral fatigue evidenced by a reduction of the MVIC. In contrast, only Dm and V10 of RF showed external responsiveness. Therefore, our study illustrates that the use of only internal or external responsiveness may lead to incomplete conclusions (*Husted et al., 2000*). In this way, professionals should use both, as recommended by Husted (*Husted et al., 2000*).

This study showed that Dm and V10 of RF measured by TMG were both internally and externally responsive to changes between before and after a peripheral fatigue protocol. Since the QF is the main strength contributor during cycling (*Raasch et al., 1997*) or running (*Montgomery, Pink & Perry, 1994*), the fatigue evaluation after an effort is essential to manage recovery of the athlete and the intensity of subsequent training sessions. Thus, clinicians and trainers should be able to direct the fatigue evaluations without making new efforts with TMG, taking into consideration Dm and V10 parameters in RF to discriminate the presence of peripheral fatigue and the magnitude of the strength changes and, in this way, be able to regulate training loads (e.g., in the presence of peripheral fatigue, decrease intensity or activities that involve the QF).

CONCLUSIONS

According to the results, it can be concluded about positive responsiveness of the TMG in peripheral fatigue of the QF, demonstrating that the Dm and V10 parameters of the RF present acceptable responsiveness to fatigue. Therefore, by using the TMG, it is possible to determine whether the QF shows peripheral fatigue or not, and to relate changes in the parameters with the reduction of strength. Thus, clinicians and trainers should be able to direct the fatigue evaluations without making new efforts with TMG, facilitating the regulation of training loads. Finally, future studies should examine the responsiveness of TMG to other types of fatigue and in other sports.

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ADDITIONAL INFORMATION AND DECLARATIONS

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Competing Interests

The authors declare there are no competing interests.

Author Contributions

- Rodrigo Martín-San Agustín conceived and designed the experiments, performed the experiments, analyzed the data, authored or reviewed drafts of the paper, and approved the final draft.
- Francesc Medina-Mirapeix conceived and designed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, and approved the final draft.

- José Casaña-Granell and Josep C. Benítez-Martínez conceived and designed the experiments, performed the experiments, authored or reviewed drafts of the paper, and approved the final draft.
- José A. García-Vidal analyzed the data, prepared figures and/or tables, and approved the final draft.
- Carmen Lillo-Navarro analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, and approved the final draft.

Human Ethics

The following information was supplied relating to ethical approvals (i.e., approving body and any reference numbers):

The University of Valencia granted ethical approval to carry out the study within its facilities (Ethical Application Ref: H1523633864097).

Data Availability

The following information was supplied regarding data availability:

The raw measurements are available in the Supplemental Files.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.8674#supplemental-information.

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